

Historic, Archive Document

Do not assume content reflects current scientific knowledge, policies, or practices.

19 575-1426

REPORT OF THE
PUERTO RICO
EXPERIMENT STATION
1940



U.S. DEPARTMENT OF AGRICULTURE
OFFICE OF EXPERIMENT STATIONS

PUERTO RICO EXPERIMENT STATION

of the

UNITED STATES DEPARTMENT OF AGRICULTURE

MAYAGUEZ, PUERTO RICO

**REPORT OF THE
PUERTO RICO EXPERIMENT STATION
1940**



Issued 1941



**UNITED STATES DEPARTMENT OF AGRICULTURE
OFFICE OF EXPERIMENT STATIONS**

PUERTO RICO EXPERIMENT STATION

Administered by the Office of Experiment Stations
United States Department of Agriculture

JAMES T. JARDINE, *Chief, Office of Experiment Stations*

STATION STAFF

Atherton Lee, *Director*
W. K. Bailey, *Horticulturist*
C. L. Horn, *Associate Horticulturist*
K. A. Bartlett, *Entomologist*
H. K. Plank, *Associate Entomologist*
R. H. Moore, *Associate Plant Physiologist*
J. K. Alvis, *Assistant Agricultural Engineer*
P. A. Folch, *Junior Agricultural Engineer*
J. O. Carrero, *Assistant Chemist*
Noemí G. Arrillaga, *Junior Chemist*
Francisca E. Arana, *Junior Chemist*
Merriam A. Jones, *Junior Chemist*
Howard T. Love, *Junior Chemist*
A. G. Kevorkian, *Assistant Plant Pathologist and Physiologist*
William Pennock, *Junior Agronomist*
A. R. Villamil, *Junior Agronomist*
E. Hernández Medina, *Junior Agronomist*
Beverly T. Taylor, *Scientific Aide*
Armando Arroyo, *Scientific Aide*
G. F. Anton, *Collaborator*
Astor González, *Librarian*
Violeta Biaggi, *Assistant Librarian*
C. Alemar, *Principal Clerk*
M. Hollis Kamenberg, *Clerk*
E. Avilés Lojo, *Assistant Clerk*

On the front cover is reproduced a photograph showing one of the reservoirs lying in the plant introduction garden at the experiment station. These reservoirs impounded by simple earth dams at low cost serve three purposes, irrigation during the dry season, flood control during the rainy season, and as a source of such fresh-water fish as bluegills and catfish. The bamboos lining the banks of the reservoirs have industrial as well as landscape value.

PUERTO RICO EXPERIMENT STATION

of the

UNITED STATES DEPARTMENT OF AGRICULTURE

MAYAGUEZ, PUERTO RICO

Washington, D. C.

May 1942

REPORT OF THE PUERTO RICO EXPERIMENT STATION, 1940

CONTENTS

	Page		Page
Introduction.....	1	Plant introductions and distributions.....	53
Population in relation to land in Puerto Rico.....	3	Biological-control activities.....	65
Investigations of vanilla production and processing.....	5	Entomological investigations.....	71
Investigations of drug plants.....	18	Chemistry investigations.....	85
Coffee investigations.....	22	Agricultural engineering.....	93
Essential-oil investigations.....	23	Improvements in property.....	96
Bamboo propagation and utilization.....	33	Cooperation with other Government organizations.....	99
Investigations of insecticidal plants.....	38	Publications.....	100
Vegetable crop investigations.....	50	Changes in personnel.....	101
Sugarcane investigations.....	51	Literature cited.....	102

INTRODUCTION

The Puerto Rico Experiment Station is an outpost of the United States Department of Agriculture engaged in research pertaining to tropical agriculture. As authorized by Congress its functions consist in (1) investigations for the benefit of Puerto Rican agriculture and (2) investigations to aid the agriculture of the continental United States, utilizing the advantages of a 12-month growing season in the Tropics.

Island agricultural policies are premised on population pressure.

Figures taken from the census of 1940 indicate that Puerto Rico now has 560 people per square mile and 0.72 of an acre of arable land per person. From 75 to 90 percent of the income of the island, varying from year to year, results from export of agricultural products, that is, products of the soil. With but 0.72 of an acre of arable land per person, it is evident that to maintain reasonable standards of income and living, the land must be used for crops of high value per acre. This is the most important factor in determining types of land utilization in Puerto Rico, and all investigations of the experiment station having to do with island agriculture are premised on this factor.

Soil erosion and climatic conditions are other basic factors which influence land utilization.

Puerto Rico, with a large proportion of its land in steep hillsides, and with intense rains of large-sized drops, is especially subject to

losses from soil erosion. The selection of crops for these hillside lands which will check erosion and favor conservation of soils is another basic factor upon which the investigations of the experiment station are premised.

With an average of one hurricane every 12 years, crops that will be least damaged by strong winds must be considered. One of the greatest assets of the island is its climate, with complete freedom from frost liability and 12 months of growing temperatures. The utilization of this asset for Puerto Rican agriculture is still another basic guiding influence upon the work of the experiment station.

Investigations on drug and insecticidal plants are for benefit of continental agriculture.

The insecticidal plant project is one example of the investigations undertaken by the station for the benefit of the agriculture of the continental United States. Fruit and vegetable growers, to avoid poisonous inorganic insecticidal sprays, are using rotenone insecticides which, although toxic to cold-blooded animals such as insects, are relatively harmless to humans and domestic animals. The highest yielding rotenone plants are tropical species. The studies upon insecticidal plants have as their objective the increasing of a reliable supply and the cheapening of rotenone insecticides. Another project for the benefit of the economy of the continental United States is the development of a secure source of quinine; the work of the experiment station during the past year has put forward considerably the final achievement of this objective.

During the year several beneficial species of insects obtained by the experiment station from the Caribbean area and South America, and which prey upon or parasitize insect crop pests, were made available for colonization in the southern United States to aid in the control of crop pests there. These are but a few of the material contributions this experiment station in the Tropics can make to continental agriculture.

In figure 1 is shown the technical personnel of the experiment station. A large proportion of the personnel speaks Spanish as well as English and with knowledge of tropical crops can well serve in implementing the good-neighbor policy of the American republics.

New crop plants and beneficial insects have been made available to neighboring republics.

The collection of economic plants at the experiment station, said to be the largest collection of tropical economic plants in the Western Hemisphere, contains many crop species from the Eastern Hemisphere difficult to obtain by an individual or private corporation, but which the United States Department of Agriculture has been able to secure through its plant exploration service aided by its quarantine service.

Because of this collection it has been possible for the experiment station to be of service to neighboring republics by making some valuable economic crop plants available for establishment, aiming toward commercial production. During the past year, for example, the station has made available seeds of teak, Venezuelan mahogany, and new and improved mango varieties, also propagating material of new industrial bamboos, insecticidal plants, cinchona and other drug plants, essential oil plants, fiber plants, and other minor crops.

Using the widespread activities and organization of the Department of Agriculture, beneficial insect species which parasitize crop pests have been assembled from many foreign countries at the experiment station and are now being made available for colonization in some of the neighboring Western Hemisphere republics.

POPULATION IN RELATION TO LAND IN PUERTO RICO

Acreage of arable land increased in past 5 years.

From the new data on population and land in Puerto Rico recently made available by the Bureau of the Census, it is evident that the acreage of arable land in the island is now slightly greater than it was in 1930, and considerably greater than in 1935, as shown in table 1. From 1910 to date, with the exception of the census year 1935, there have not been great variations in the acreage of arable land



FIGURE 1.—The technical personnel of the Puerto Rico Experiment Station of the United States Department of Agriculture.

in the island. It seems probable that the low figure in 1935 may have been a result of the low prices for commodities which prevailed during the economic depression; much of the land withdrawn from cultivation in that period could easily have been classed as nonarable.

TABLE 1.—*Figures from the United States Bureau of the Census showing population and areas of arable land in Puerto Rico for the census years 1899 to 1940*

Census year	Population	Population increase, 10-year period	Average members per family	Total arable land	Arable land per person
		Percent	Number	Acres	Acres
1899	953, 243		5. 2		
1910	1, 118, 012	16	5. 1	1, 570, 304	1. 40
1920	1, 299, 809	16	5. 2	1, 303, 547	1. 00
1930	1, 543, 913	19	5. 3	1, 222, 284	. 79
1935	1, 723, 534		5. 4	827, 350	. 48
1940	1, 869, 245	21	5. 3	1, 341, 938	. 72

Area of arable land per person is now 0.72 acre.

The population of Puerto Rico, according to the census of 1940, was 1,869,245, an increase of 21 per cent as compared with 1930. With the 1,341,938 acres of arable land during the same census year, the average tillable area per person is now 0.72 of an acre. With an average of 5.3 individuals per family, the average amount of arable land per family at the present time is 3.82 acres.

Areas of cultivable land can be increased.

Familiarity with the topography and climatic conditions in Puerto Rico makes it obvious that greater areas of land can be made productive. It is evident that areas of marshy land, in some cases saline, are being and can still further be recovered by drainage and leaching. The percentage increase in arable land by this method, however, would not be expected to be great.

In the southwestern part of the island, in the regions of low average annual rainfall, there are areas of semiarid lands which could be made more productive with irrigation. A large proportion of these lands is now idle or utilized as poor pasture with almost negligible income.

The greatest increase in acreage of cropland probably can be obtained by adapting new crops to some of our hillside conditions. Experience has shown that such tree crops as mangoes can be produced to advantage on 20-percent or greater slopes. Other tree crops for which experience has now accumulated and which are adapted to our hillside lands in the varied rainfall areas of Puerto Rico are papayas, avocados, the sapote and sapodilla, ilang-ilang, *Aleurites trisperma* Blanco, a tropical tung-oil species, and bamboos. Vanilla is another crop well adapted to steep hillsides. Sugarcane could be extended to moderately sloping lands, thus making available greater areas of land for the production of winter vegetables which could be expected to yield higher gross income per acre than sugar. Sugarcane planted in contours properly timed is a good crop to avoid soil erosion.

It is conceivable that by these means areas of cultivable land in the island can be increased by several hundred thousand acres.

Population almost doubled in past 40 years.

Table 1 shows an increase in population of 916,002, or 96 percent, in the 41 years from 1899 and 1940. These figures have acute significance in view of the difficulty of sustaining even the present population with the present limited area of arable land.

There is also perhaps some significance in the figures showing the rates of population increase during the different decades between census enumeration. The rate of population increase has augmented considerably from 16 percent in the decade ending in 1910 reaching to 21 percent in the decade ending in 1940.

During the 36 years from 1899 to 1935 for which census figures are now available, there was a steady increase in the average number of individuals per family, 5.1 in 1910 to 5.4 per family in the census figures for 1935.

It seems a logical conclusion that well-studied emigration projects are needed to supplement agricultural and economic developments to improve or even maintain the present island economy.

INVESTIGATIONS OF VANILLA PRODUCTION AND PROCESSING

PRODUCTION STUDIES

Root rot has been limiting factor in vanilla production in Puerto Rico.

Puerto Rico, at a latitude but slightly closer to the equator than the native habitat of vanilla in Mexico, has seemed to have conditions well adapted for this crop. Vanilla cultivation yields high gross income per acre which, with the crowded population and limited land areas in the island, is an essential for any new crop under consideration. There is at present a tax of 15 cents per pound on vanilla beans entering the United States from foreign countries, which gives the island a small price advantage.

Vanilla has a high value per unit of weight, and consequently the cost of transportation is relatively small; it is therefore well adapted to cultivation on steep hillsides from which it is difficult to harvest and transport other crops. Vanilla, with its shade trees, also prevents soil erosion. Thus vanilla production has advantages in the island from the standpoint of climate, economics, and topography.

The limiting factor to expansion of this crop to the present time has been a fungus disease of the roots, described in previous reports, which has seriously limited production. In some plantings as many as 75 percent of the plants have been eliminated from production by this disease.

Disease is caused by a fungus of the genus *Fusarium*.

Work at the experiment station by Tucker (14)¹ in 1927 showed that the casual fungus of vanilla root rot is *Fusarium batatatis* var. *vanillae* Tucker. Root rot of other crop plants caused by fungi of this genus are well known and of great economic importance throughout the world, particularly in the warmer countries. At least several of the pathogenic species of this fungus are soil-inhabiting. In the case of the other crop plants, diseases caused by species of *Fusarium* are favored by heavy, wet soils, and drainage and good aeration are usually recommended as one means of minimizing them.

Two districts in Puerto Rico have no commercial losses from root rot.

Vanilla has been grown in the Morovis-Ciales district of Puerto Rico since 1921 without serious losses from root rot. The soils in this district are of limestone origin, neutral or slightly alkaline in reaction, and characterized by good aeration and drainage. On the Cerro de las Mesas in the western part of the island, vanilla has been grown for 4 years with a minimum of losses from root rot. These soils are of serpentine origin entirely distinct geologically from the soils of the Morovis-Ciales district. The Las Mesas soils, however, are similarly characterized by a neutral or slightly alkaline reaction and aeration and drainage are unusually good.

Terrestrial orchids need well aerated soils.

In nature throughout Central and South America, terrestrial orchids are found on rocky, steep hillsides or frequently in gravelly

¹ Italic numbers in parentheses refer to Literature cited, p. 102.

areas of lesser slopes. Other species of the genus *Vanilla* occurring naturally are usually found in well drained and aerated soil environments. The Orchidaceae in general, being epiphytes, require a high degree of aeration for their roots.

Root rot at the experiment station has occurred on heavy, highly compact clay soils of high water-holding capacity.

It seemed from the foregoing background that experiments to produce vanilla in soils of better aeration and with closer attention to soil reaction might produce an environment more favorable for the vanilla plant and less favorable for the root rot fungus.

Tests showed poor results from mulches of charcoal, coconut husks, and coffee pulp.

In an attempt to provide a more favorable environment for vanilla roots, charcoal, coconut husks, and coffee pulp were tested as mulch substrata in comparison with ordinary Catalina clay found so extensively in the vanilla-growing districts of the island. Large percentages of root rot were obtained in all treatments as well as in the controls.

In a second experiment, in which different proportions of gravel were incorporated with leaf mulch to provide greater aeration for the roots, only small percentages of root rot were obtained. The experiment showed few differences in percentages of root rot between treatments, apparently because the mulch became compacted and the plants were so close together that but small experimental differences could be obtained. However, it was outstanding that throughout the experiment in which gravel was used there was but a small percentage of the disease, whereas in the contiguous experiment, testing the heavier, water-holding mulches without gravel, 75 percent of the plants were affected. One entering the experimental shaded area was immediately impressed by the difference in incidence of root rot. These results are shown in figures 2 and 3.

A somewhat similar experiment is now being carried on, profiting from the experience showing the deficiencies in the nature of the previous experiment. It was outstanding from the original experiments, however, that gravel introduced into the substratum minimized root rot.

Expansion of vanilla cultivation in known regions of little root rot is logical.

In a representative year the consumption of vanilla in the United States is about 1,200,000 pounds, or 600 short tons. The area required to produce this quantity, conservatively, need not be more than 20,000 acres. There is sufficient area, in the limestone hills in the north-western part of Puerto Rico and in the soils of serpentine origin in the west and southwest, to produce sufficient vanilla to meet the requirements of the United States at present without attempting to grow the plant in the regions of high root rot incidence. It would seem a logical conclusion, therefore, that attempts at expansion of vanilla production should be confined to these regions of low root rot incidence while continuing experiments on the environmental conditions which favor the absence of root rot.

A blast disease of vanilla flowers and fruits was observed.

A vanilla disease that attacks undeveloped flower buds and immature pods and causes them to drop was mentioned in the annual re-



FIGURE 2.—Vanilla plants grown on heavy mulches of high water-holding capacity; the flaccid, wilting leaves typical of root rot indicate the large percentage of plants affected by the disease.

port for 1939. This disease, caused by an undetermined fungus, appears to have become more widespread during the time it has been

under observation. It has become established in several vanilleries in Mayaguez and at Morovis, as well as at Villalba, where it was first



FIGURE 3.—Vanilla plants grown on various mixtures of leaf mulch with gravel, producing good aeration and drainage in the substratum; these plants were mostly healthy at the termination of the experiment. Note turgor of the leaves of the plants as compared with the flaccidity of the leaves in figure 2.

discovered. In the last-named location the infection in the 250 plants under observation increased from 31 percent in 1939 to 44.8 percent

during the past year, an increase of 13.8 percent in number and 44.5 percent in proportion of plants infected.

Presence of the disease coincided with extent of shade.

The losses due to this disease, as might be expected, varied with climatic and environmental conditions. In one locality at Villalba, where the shade was slightly denser, 68 percent of the plants were infected to some extent, whereas in another area slightly less shaded there was but 8 percent infection. Undoubtedly, the more shaded area was more humid during the critical period of the fungus development and conditions were thus more favorable for its spread.

Multiple flower clustering was associated with the disease.

In some plants the necrotic area was confined to the apical portion of the cluster. In such cases the primary cluster gave rise to secondary flower clusters from the basal bracts of the peduncle of the original cluster. These secondary clusters produced flowers, and when pollinated, gave rise to normal pods. Multiple clusters have been found to contain from 20 to 30 pods. Occasionally a healthy-appearing multiple cluster may be seen, as shown in figure 4, but clusters either adjacent to it or at a distance on the same vine will be infected. This proliferation of flower clusters has not been observed in plants free from infection.

Hold-fast roots showed little capacity to utilize nutrients.

The vanilla plant puts out hold-fast roots at each node which serve to attach it firmly to the support tree. It is always a question in the minds of vanilla growers and investigators as to what extent these hold-fast roots serve in the utilization of nutrients. An experiment was therefore undertaken during the year in which vanilla vines were grown under identical conditions, with the variable introduced of different types of support. Some plants were grown on living stakes of *Erythrina berteriana* Urban, others on dead stakes of the same species, others on well-seasoned stakes of dead hardwood from which the bark had been removed, and a fourth group on simple cotton cords suspended from the shade supports above. There were two replications of each treatment with four plants in each replication, so that the experiment can only be considered as one of orientation. Table 2 shows the average linear growth per plant in each treatment and the total weight of all plants per treatment at the end of 12 months.

TABLE 2.—Showing longitudinal growth per plant and weights of vanilla vines growing on various types of support

Form of support	Longitudinal growth per plant	Weight of vines per treatment
	Feet	Pounds
Living <i>Erythrina</i>	15.5	10.75
Dead <i>Erythrina</i>	14.8	8.50
Dead hardwood stakes without bark.....	17.7	11.50
Cotton cord.....	19.6	12.25

It can be seen from table 2 that although the results were not necessarily statistically significant, the plants supported by the cotton cord outgrew the plants upon all classes of support stakes. It may be concluded from this experiment, therefore, that since the plants on cotton cord with no nutrients outgrew the plants on support



FIGURE 4.—Abnormal flower cluster of *Vanilla fragrans* (Salisb.) Ames. Note the proliferation from the basal bracts of the peduncle. This abnormality has only been found associated with the diseased condition causing blossom blast and fruit rot. The adjacent flower cluster on this vine also showed the diseased condition.

stakes containing some degree of nutrients, the hold-fast roots did not function greatly in the intake of plant foods. Or, stated in another way, most of the nutrients were obtained by the vanilla plants through their terrestrial roots.

These conclusions are supported by a case in which a vanilla vine in one of the other experiments escaped from its support stake and attached itself to an iron trellis support. The longitudinal growth of the vine on the iron tube was fully as good as the growth of any of those on the wooden support stakes. It is conceivable, of course, that support material will be obtained in the future from which hold-fast roots might obtain some nutrients, but it is apparent that vanilla can grow vigorously without intake of nutrients through the hold-fast roots.

PROCESSING STUDIES

Moisture content of beans can be controlled in processing.

In experimentation in previous years a contribution has been made to the processing of vanilla beans by substituting the uniform temperatures of electric heating devices, controlled by thermostats, for the irregular heat of the sun-curing process; in the last-mentioned process, beans exposed to the sun, at night are wrapped in blankets to hold the heat. The exact temperatures to which the beans are being submitted, in the sun or when wrapped in blankets, is not known. As a result of the work this year it has been found that the peroxidase activity in the beans is retarded by exposure to sun. Since these studies have established that the curing of vanilla beans is largely a process depending on enzyme activity, the substitution of heat chambers of uniform known temperatures, favorable to the specific enzymes concerned, have considerably aided such processes.

Another material contribution has been made in achieving control of moisture losses during processing. The beans were weighed before killing was undertaken, and thereafter at the end of each processing step weights of the beans were determined. The effect of different killing treatments on moisture content of the beans was determined, and, particularly in the sweating chambers and drying racks, it was possible to modify moisture content of the end product.

Excessive drying limits enzyme activity.

In these experiments weight of the beans was reduced in the sweating and drying stages by 55, 65, and 75 percent of the original weight. It was clearly evident that those beans cured to 45 percent of their original weight, although having the best appearance and greater flexibility, were subject to attacks of molds. The processing in which the beans were reduced to 35 percent of their original weight resulted in the retention of much of the favorable qualities of the beans of higher moisture content, and the beans evidenced heavy formation of vanillin crystals, shown in figure 5, as would be expected in a medium sufficiently moist for enzyme activity to continue. In the processing in which the beans were reduced to 25 percent of their original weight, the ends became hard and somewhat woody and the beans had lost the flexibility and pleasing appearance of those of higher moisture content; the best aroma, however, was obtained in these beans.

It was found that the greater the degree of drying of the beans, the less their respiration following conditioning, as evidenced by carbon dioxide evolution; this would be naturally expected since



FIGURE 5.—Vanilla beans showing heavy formation of vanillin crystals when killed by water at 80° C. and reduced to 35 percent of their original weight.

the greater moisture content would favor greater enzyme activity. The evidence at present points to maintaining the moisture content

of the beans as high as possible during curing and conditioning, before drying to the moisture percentage at which sales will be made. Obviously the moisture content must be maintained below the point at which molding will occur.

Length of exposure to killing agents governed splitting of beans.

Split beans frequently yield lower prices than whole beans. From these experiments it became evident that the shorter the exposure of the beans to the killing agent, the less splitting would occur. As an example, in comparative experiments in which beans of the same degree of maturity were exposed to sunlight, freezing, hot-water treatments, and ethylene gas, splitting was the greatest in those beans exposed to sunlight intermittently for several days. Splitting was least when the beans were exposed to hot-water treatments of but a few seconds. In experiments to test exposures to ethylene gas, splitting was always greatest in those treatments having the longest time of exposure. Similarly, splitting was greater under long exposure to freezing temperatures than with short periods of exposure. The conclusion was rather definite that some control could be exercised over the splitting of the beans by shortening the time during the killing and to a lesser degree by shortening the time of the whole curing process.

Dilute concentrations of ethylene gas gave better results than greater concentrations.

In previous years favorable results have been obtained by using ethylene gas in the killing or first stage of processing of vanilla beans. During the past season experiments were conducted to test the concentrations and optimum periods of exposure of the beans to ethylene gas. It was found that 1 part of the gas to 500 parts of air was in most cases detrimental in that less crystals, less pleasing aroma, and poorest color were obtained. At prolonged exposures of 16 or 32 hours a 1-1,000 concentration of the gas was also detrimental. Dilutions of 1-100,000 gave better results for an exposure of 8 hours as regards vanillin, total solids, and resin content than for exposures of 4, 12, or 16 hours. These tests showed that ethylene gas produced the best aroma in green beans and the poorest aroma in the more mature blossom-end-yellow or chocolate-colored beans. The exposures were all made at normal mean indoor temperatures of from 73.3° to 74° F. during the months of January to April.

CHEMISTRY OF VANILLA PROCESSING

Vanilla beans have high content of peroxidase.

In the report for 1939 mention was made of tests showing the presence of the enzymes, oxidase and peroxidase, in vanilla beans. In the year under review, quantitative tests of peroxidase in vanilla beans were made. A conception of the peroxidase content of green and cured vanilla beans as compared with other vegetable tissues notable for their peroxidase content is shown in table 3. It can be seen that the peroxidase content of vanilla beans is considerably higher than that of wheat, and is more comparable to that of potatoes. Apparently little peroxidase content was lost during the processing in this case, inasmuch as the cured beans showed almost as great peroxidase activity as the green vanilla beans.

TABLE 3.—*Relative peroxidase content of certain plant tissues, including vanilla*

Plant tissues	Peroxidase content per kilogram at 30° C., dry basis
	<i>Peroxidase units</i>
Malt sprouts ¹	550
Horseradish ¹	1,160
Turnips ¹	1,100
Potatoes ¹	180
Wheat ¹	10
Green vanilla beans	(A) 150
Do.	(B) 190
Do.	(C) 140
Cured vanilla beans	(A) 120
Do.	(B) 160
Do.	(C) 130
Seeds and surrounding tissues from cured vanilla beans	21

¹ These values, taken from Balls and Hale (3, p. 452), were calculated to approximate dry basis for this table.

The peroxidase activity of beans subjected to different methods of treatment was also determined. Table 4 shows the peroxidase content of two lots of beans after such various methods of processing. It is notable that killing of vanilla beans by hot water or exposure to ethylene gas markedly increased peroxidase activity. On the other hand, killing of vanilla beans by freezing for 16 hours greatly reduced it. Peroxidase activity was not significantly altered by exposure to the sun for 5 hours or in an electric oven at 50° C. for 16 hours. During the 24 to 26 days of conditioning following killing, there was a notable drop of peroxidase activity for all treatments.

All lots with the exception of lot M-2 were treated identically after the original killing procedures.

TABLE 4.—*Peroxidase activity of vanilla beans during the curing process*

Sample or lot No.	Method of killing	Peroxidase content per kilogram of dry weight		
		Fresh beans	After killing	Twenty-fourth to twenty-sixth day of conditioning period
		<i>Peroxidase units</i>	<i>Peroxidase units</i>	<i>Peroxidase units</i>
A-1	Oven, 50° C. for 16 hours	116	114	
M-1	do.	126	107	92
A-2	Sun, for 5 hours	116	107	
M-2	do.	126	¹ 135	² 80
A-3	Ethylene (1-10,000) 16 hours	116	160	
M-3	do.	126	149	79
A-4	Frozen for 16 hours	116	53	
M-4	do.	126	107	67
A-5	Dipped thrice for 10 sec. in water at 80° C	116	162	
M-5	do.	126	142	70

¹ Sunlight was weak during the wilting period.

² These beans were sweated in the sun (Mexican method) for 15 days, and at the time of the analysis had been kept only 14 days in the conditioning trunk.

Balls and Hale (4, p. 778) have shown that vanillin is acted upon by the peroxidase obtained from horseradish. In the experiment station laboratories during the past year it was shown that the peroxidase from vanilla beans similarly acted upon vanillin. Moreover, it was shown that the rate of oxidation produced by peroxidase on vanillin was closely similar to its rate of oxidation on pyrogallol. Similar to the results of Balls and Hale with the peroxidase of horseradish were results showing that the peroxidase of vanilla beans has little effect on phloroglucinol and resorcinol. The evidence in the laboratory to date, therefore, is that the peroxidase of vanilla beans is closely comparable to that of horseradish.

Tests made for peroxides in cured beans indicated their presence. It is therefore evident that a complete peroxidase enzyme system is present in such beans, with the vanillin as the substrate acted upon by the oxygen released from peroxides by the peroxidase. There is therefore some basis for the inference that vanillin in vanilla beans serves not only in itself to produce aroma and flavor but also to produce more elaborate aromatic and flavoring compounds.

Ethylene gas, as killing agent for vanilla beans, increased respiration.

In previous years it has been shown that ethylene gas used as a killing agent for vanilla beans materially accelerated processing. Inasmuch as studies of other fruits, such as citrus, bananas, and tomatoes, have shown that an increased rate of respiration accompanies the accelerated maturity, it seemed logical to undertake measurement of rates of respiration in the cases of vanilla beans subjected to different killing agents. The rate of respiration immediately following killing was determined by measurement of carbon dioxide evolution. The rates of evolution of carbon dioxide from vanilla beans subjected to various killing treatments are given in table 5, which shows that ethylene gas materially increased the rate of carbon dioxide evolution in vanilla beans. Scratching the beans with a pin, which in effect would be to rupture the epidermis, increased the rate of carbon dioxide evolution even more than the ethylene treatment. All of the treatments used in the foregoing experiment increased materially the rate of carbon dioxide evolution.

TABLE 5.—Rate of carbon dioxide evolution by green vanilla beans subjected to skin injury

Killing treatment	Carbon dioxide respired per hour per kilogram of—	
	Treated beans	Control beans
	Milligrams	Milligrams
Exposure for 16 hours to ethylene gas, 1-10,000	102	66
Rubbed with 50-percent alcohol	58	47
Naturally split beans	97	46
Dipped in hot water ¹	81	47
Scratched with a pin	165	86

¹ Immersed 3 times for 10 seconds at 30-second intervals in water at 80° C. It is obviously impossible to heat the beans through by this treatment.

Freezing was used as method to decrease respiration of vanilla beans.

Since in all of the foregoing treatments of vanilla beans the rates of respiration were increased, it seemed desirable to test the effect of some killing method which would decrease the respiration rate. Experiments with freezing as a means of killing the beans were therefore undertaken. Its effect upon the respiration of the beans is shown in table 6, from which it will be noted that freezing of the beans materially lessened the evolution of carbon dioxide. The cured beans following the freezing treatment had a reddish-brown color and were considerably more flexible than those treated with ethylene gas or hot water. Moreover, this flexibility in the case of the frozen beans extended to the tips of the beans, which, following other methods of treatment, usually are hard and woody and are not considered of great value for extraction.

TABLE 6.—Rate of carbon dioxide evolution by green vanilla beans subjected to freezing

Treatment time of freezing (hours)	Period after thawing when readings were taken	Carbon dioxide respired per hour per kilogram of—	
		Frozen beans	Control beans
	Hours	Milligrams	Milligrams
16.....	3	27	42
24.....	2	14	47
48.....	2	13	47

Killing agents affected respiration rate during whole processing period.

To substantiate further the foregoing experiment, carbon dioxide evolution was determined throughout all stages of processing following a 16-hour exposure to ethylene gas of green beans as compared with a hot-water treatment of mature beans and a freezing treatment also of mature beans; the results are shown in table 7.

TABLE 7.—Rate of carbon dioxide evolution during the curing process of vanilla beans subjected to three killing treatments

Lot No.	Maturity of beans	Killing treatment	Carbon dioxide evolved per hour per kilo-gram of beans—							
			Before treatment	1 hour after treat-ment	During sweat-ing			At end of sweating and drying	During condition-ing	
					1 day	3 days	6 days		15 days	8 months
1	Green	16-hour exposure to ethylene, 1-10,000	Mg.	Mg.	Mg.	Mg.	Mg.	Mg.	Mg.	Mg.
2	Yellow	Dipped thrice for 10 seconds in water at 80° C	66	102	100	57	63	43	15.0	9.6
3	do	Frozen 16 hours	79	123	111	109	28	21	6.0	7.1
			79	73		64	60		12.8	4.9

The ethylene treatment increased the rate of respiration, and such respiration continued actively throughout the sweating and drying stages of processing. Treatment with hot water accelerated the respiration even to a greater extent than ethylene gas, but such respiration decreased more rapidly than in the case of ethylene treatment. This difference in respiration rates between these two treatments, however, may have been due to differences in the degrees of maturity of the beans under treatment. Following the freezing treatment, respiration was immediately decreased; but the carbon dioxide evolution was maintained approximately constant for a longer time than that of the hot-water and ethylene treatments. During conditioning carbon dioxide evolution became almost negligible in all treatments.

Experiments are now going forward to test the effect of decreased respiration as compared with increased respiration on the aromatic and flavoring qualities of the bean.

Various degrees of maturity were studied in relation to processing.

In previous years it had become apparent that the degrees of maturity of vanilla beans before initiating processing had considerable influence on the quality of the end product. For the studies in the year just completed four classes of maturity of beans were selected. The most mature beans found in vanilleries are those which have turned chocolate color. The next most advanced degree is that in which the beans have turned a yellow color at the blossom end and have split longitudinally. Beans the blossom ends of which have turned yellow but which have not split are classed as next advanced in maturity. Green beans with no yellow coloring are considered least advanced toward maturity.

In the studies it was shown that green beans, after curing, gave the blackest end product. Beans which had their blossom ends yellow or were chocolate-colored yielded a reddish-brown end product.

When these beans of various degrees of maturity were carried through to extraction it was found that the chocolate-colored beans gave the darkest-colored extract, whereas the black cured beans which resulted from the green beans of least degree of maturity gave an extract of a lighter brown color.

Vanillin content and total solids values were higher in extracts of mature beans.

Analyses of the extracts of beans from different classes of maturity processed identically showed that the vanillin content and total solids values were generally higher in the whole and split beans with yellow blossom ends. In most cases the chocolate-colored beans at the completion of processing had lower content of vanillin and total solids than the beans with blossom ends yellow, this being possibly due to physical losses since the more mature beans have a greater proportion of splitting. The low vanillin content might also possibly have been due to its more advanced oxidation. This conception confirms the experience at the station that aging of the cured beans has a definite value. The green beans had the lowest contents of vanillin and total solids at the completion of processing.

As regards resin contents, the chocolate-colored beans following processing made the best showing in two out of three comparisons.

Standardized organoleptic tests were developed to measure value of cured beans.

In the curing experiments of the past several years it has become evident that analyses for vanillin, total solids, and resins do not as yet evidence the value of vanilla beans or the aroma and flavor of the extracts. To an even lesser extent do the physical characteristics of the beans, such as color, flexibility, or crystal formation, indicate such value for extract purposes. It therefore became evident that some standardization of organoleptic tests would be necessary in evaluating the end products of various experiments on the maturities of beans or killing or conditioning treatments. At the experiment station such organoleptic tests were developed for the cured beans, and samples of extracts were sent to the Bureau of Home Economics of the Department of Agriculture where more elaborate tests of the extracts were developed.

Organoleptic tests of extracts by the Department agreed well with similar tests of cured beans in Mayaguez.

In the organoleptic comparisons of cured beans at the experiment station, samples were given lettered designations and presented to a committee of technologists familiar with the vanilla work for grading as to aroma and visible presence of vanillin crystals.

In the Department tests, fresh milk was selected as the best medium for a comparison of different extracts, and controls of ethyl alcohol of the same concentration as that used in the vanilla extract were used in fresh milk as a basis of comparison. Each sample was judged four times by each of seven persons, who then ranked the samples according to preference. The extracts were judged for intensity and desirability of flavor and aroma. It is apparent that at this stage of knowledge of the chemistry of vanilla such standardized organoleptic tests are the best method of evaluating vanilla beans.

It is interesting to observe that the organoleptic tests conducted by the committee of the Bureau of Home Economics on the extracts tested in milk agreed generally with the evaluations given to the cured beans in organoleptic tests by the committee at the experiment station.

Appreciation is expressed to Florance B. King, in charge, Food Utilization Section of the Bureau of Home Economics, for the assistance rendered in her standardization studies for evaluating vanilla extracts.

The studies of the chemistry of vanilla have been carried on by Francisca E. Arana, junior chemist. During the year A. K. Ball's, head chemist, Enzyme Research Laboratory of the Bureau of Agricultural Chemistry and Engineering, participated in our laboratories in the studies of the chemistry of vanilla processing. Appreciation is expressed to Dr. Balls for his close collaboration from which resulted much of the progress achieved in these studies.

The investigations of vanilla production and processing have been carried on by Arthur G. Kevorkian, assistant plant pathologist and physiologist, and Ernesto Hernández Medina, junior agronomist.

INVESTIGATIONS OF DRUG PLANTS

Cinchona seeds lose viability rapidly.

In a study of storage of seeds of *Cinchona officinalis* L. in which more than 6,000 seedlings were grown, it became evident that there

was a rapid falling off in viability with increasing age of seed under usual atmospheric conditions in Mayaguez. In general, seed older than 186 days, or approximately 6 months old at the time of planting, germinated poorly. Seed more than 250 days, or approximately 8 months, old had almost completely lost its viability.

The studies also indicated that *Cinchona* seeds germinate slowly. The data in table 8 illustrate the slowness of germination of seeds of *C. officinalis*.

TABLE 8.—Cumulative counts of germination of four lots of seed of *Cinchona officinalis* 49 days old at time of planting

Tree No.	Lot No.	Seeds germinated in—				
		21 days	30 days	56 days	63 days	103 days ¹
38.....	1	Number 8	Number 70	Number 200	Number 230	Number 268
15.....	2	35	180	440	450	500
111.....	3	200	440	750	766	775
111.....	4	85	265	510	525	600
Total.....		328	955	1,900	1,971	2,143
Proportion.....		Percent 15.30	Percent 44.56	Percent 88.66	Percent 91.97	Percent 100.00

¹ Further germination was not observed to occur beyond this period.

Cinchona seedlings were highly sensitive to modifications of light intensity.

Cinchona seedlings were found to be highly sensitive to slight modifications in light intensity. Good results were obtained when the seeds were germinated under heavy shade that excluded almost all light and where, following germination, the shade was thinned out progressively as the seedlings increased in size. When the seedlings attained a height of 4 to 6 inches, the shade was reduced so that the light entering registered about 35 foot-candles on a Weston photometer when full sunlight registered 12,000 foot-candles.

It was observed that *Cinchona* seedlings exposed to a slight excess of light became weak and developed a reddish color, while with more severe overexposure leaf burning occurred. In slightly excessive shade, growth of the *Cinchona* seedlings was delayed; in greater excess of shade undesirable etiolation occurred. Seed germinated best in the dark, and small young seedlings were more sensitive to injury from excess light than the larger and older seedlings.

William Pennock, junior agronomist, was in charge of cinchona production studies during the year. At the close of the year he had accepted a position as manager of the *Cinchona* plantations of Merck & Co., Inc., in Guatemala.

Studies were undertaken to standardize methods of determining quinine in bark.

In studies to standardize methods of quinine determination, ether-chloroform extraction of the bark was compared with continuous extraction with ammoniacal alcohol. The ether-chloroform method gave more consistent results; it was thereafter considerably standardized. The great similarity in the molecular structure of the four

main cinchona alkaloids so important commercially, namely, quinine, cinchonidine, cinchonine, and quinidine, and their correspondingly similar reactions to solvents and other chemical reagents, makes their isolation difficult and tedious.

Quinine content of bark varied widely in same tree.

It was shown that the alkaloid contents of cinchona bark varied greatly not only from tree to tree but in different parts of the same tree. The variation in the quinine content of the bark in different parts of an individual *Cinchona* tree can be quickly appreciated by reference to tables 9 and 10.

TABLE 9.—Analysis of bark taken from four sides of three 7-year-old *Cinchona officinalis* trees in the Maricao planting

Side of tree	Tree No. 107 ¹		Tree No. 108 ²		Tree No. 111 ³	
	Quinine sulfate	Total alkaloids	Quinine sulfate	Total alkaloids	Quinine sulfate	Total alkaloids
	Percent	Percent	Percent	Percent	Percent	Percent
Downhill.....	6.9	8.7	6.3	6.8	7.4	10.3
Left.....	5.9	7.5	9.5	11.9
Uphill.....	5.2	7.9	5.1	5.8	10.9	13.5
Right.....	6.4	8.2	9.3	11.8

¹ Tree No. 107, 9 feet high, was in a poor condition of health and was in flower at time of sampling. It had a poor root system and few branches, and the trunk was cankered on the uphill side.

² Tree No. 108, 14.5 feet high, was in fair condition of health and was in flower. There was good lateral distribution of branches with a trend towards downhill branching.

³ Tree No. 111, 15 feet high, was in poor health and not in flower. There was poor distribution of branches. Most of the root system appeared to be on the uphill side.

TABLE 10.—Longitudinal distribution of alkaloids in bark of three 7-year-old *Cinchona officinalis* trees in the Maricao planting

TREE NO. 107¹

Sample No.	Height from ground	Circumference of tree	Quinine sulfate	Total alkaloids
	Feet	Inches	Percent	Percent
1.....	0.5-1.0.....	6.75	6.2	8.8
2.....	1.5-2.0.....	6.00	6.0	8.7
3.....	2.5-3.0.....	5.50	6.7	9.1
4.....	3.5-4.0.....	5.90	5.3	8.9
Average.....	6.1	8.9

TREE NO. 108¹

1.....	0.0-0.5.....	10.00	7.0	7.4
2.....	1.0-1.5.....	9.00	5.3	6.1
3.....	2.0-2.5.....	8.25	4.8	5.2
4.....	3.0-3.5.....	8.00	3.3	4.1
5.....	4.0-4.5.....	7.50	3.9	4.8
6.....	5.0-5.5.....	7.00	4.1	4.6
7.....	6.0-6.5.....	6.50	3.1	3.6
8.....	7.0-7.5.....	6.00	3.8	4.2
9.....	8.0-8.5.....	5.00	4.1	4.6
Average.....	4.4	5.0

TREE NO. 111¹

1.....	0.5-1.0.....	8.75	9.8	12.2
2.....	2.5-3.0.....	7.50	8.1	11.4
3.....	4.0-4.5.....	5.75	6.2	10.7
4.....	5.0-5.5.....	6.25	7.0	11.8
5.....	7.0-7.5.....	4.75	4.2	7.6
Average.....	7.0	10.7

¹ See footnotes below table 9 for description of these trees.

Tables 9 and 10 show that in general the bark from the lower parts of the tree contained more quinine and total alkaloids than that from the upper parts. These tables also indicate representative quinine percentages found in the bark of *Cinchona officinalis* trees in the Maricao planting of the station.



FIGURE 6.—Section of a *Cinchona* tree in the Maricao planting showing method of sampling. New bark has partially filled the peeled areas after 6 months. Most of the branches of this tree were on the downhill side.

From the foregoing variability in quinine content in different parts of the same tree, it was evident that a standardized method of sampling would be necessary. Such a method developed during the year was as follows: Starting at the base, strips of bark 6 inches long and approximately $\frac{1}{2}$ inch wide were longitudinally peeled from the trunk, as shown in figure 6. These strips of bark were alternately

taken from different sides of the tree which were designated as uphill, downhill, and, facing uphill, right and left. A portion of each of the several strips taken from the downhill side of the tree was combined to form a composite sample of the downhill side. Composite samples of the uphill, right, and left sides of the tree were obtained in the same way.

Howard T. Love, junior chemist, was in charge of studies of the chemical control and processing of drug plants during the year.

Studies of coca leaf production were discontinued.

During the past 2 years considerable success was obtained in the importation of seed of the coca plant, which produces cocaine, and the production of such plants. The environment in the western part of Puerto Rico was found to be well suited for this crop, 8,000 plants being well started and advanced in growth. However, because of the problem of controlling narcotics in the continental United States the studies of coca plants were discontinued and the plantings completely eradicated.

COFFEE INVESTIGATIONS

Columnaris variety of *Coffea arabica* L. continued to outyield West Indian variety.

In the replicated coffee variety experiment planted in 1931 on Catalina clay on the station grounds at Mayaguez by T. B. McClelland, the Columnaris variety of *Coffea arabica* L., introduced by this station from Java, continued to show a heavier yield of marketable coffee than the West Indian variety, which is most commonly grown in Puerto Rico. The yield per acre from the 1939 crop was 1,113 pounds of marketable coffee for the Columnaris variety and 1,019 pounds for the West Indian, the difference in favor of the Columnaris variety amounting to 9.2 percent. This is the sixth consecutive year that the Columnaris variety has outyielded the West Indian.

To date the Columnaris variety has produced a total of 6,861 pounds of marketable coffee per acre, while the production for the West Indian variety has amounted to only 3,915 pounds per acre. The average annual yield for the 6-year period has been 1,144 pounds per acre for Columnaris and 652 pounds per acre for West Indian. Thus, for the first 6 bearing years the Columnaris trees in this experiment have produced 75.5 percent more marketable coffee per acre than the West Indian.

Trees of Columnaris variety continued to increase in vigor.

Trees of the Columnaris variety have shown ever-increasing vigor, and although the production of the West Indian variety has been increasing progressively since 1936, it is to be expected that because of the poor vigor of the trees, yields of this variety will begin to decline sooner. The yields of marketable coffee for the two varieties for the 6-year period 1934 to 1939, inclusive, are shown by years in table 11.

TABLE 11.—*Comparative yields of the Columnaris and West Indian varieties of coffee at Mayaguez, P. R., during the 6-year period 1934-39*

Variety	Marketable coffee per acre						Six years, 1934-39	
	Crop of 1934	Crop of 1935	Crop of 1936	Crop of 1937	Crop of 1938	Crop of 1939	Total	Average
	Pounds	Pounds	Pounds	Pounds	Pounds	Pounds	Pounds	Pounds
Columnaris.....	373	1,122	1,928	1,331	994	1,113	6,861	1,144
West Indian.....	372	617	469	713	725	1,019	3,915	652

Seed and seedlings of Columnaris coffee are in great demand.

In a series of tests at the station, people who all their lives have been accustomed to using Puerto Rican-grown West Indian coffee have been unable to distinguish between coffee made from the West Indian and the Columnaris varieties. As a result of the excellent showing of the Columnaris variety in this test, seed and seedlings of this variety are in great demand by Puerto Rican coffee growers. Because of the tremendous vigor of the Columnaris trees it is likely that some modifications of the present planting, pruning, and possibly shading practices might be necessary if maximum productiveness is to be realized.

Proportion of flowers developing into mature berries varied from year to year.

For the 1939 crop 15,047 blossoms of the West Indian variety and 10,340 of Columnaris were counted and subsequent records made of the number of mature berries harvested. Of the West Indian flowers counted, 20.9 percent developed into mature berries as compared with 28.8 percent of the Columnaris flowers counted. Similar records were taken for the 1937 and 1938 crops. For these three crops the proportion of the flowers counted for the West Indian variety that set fruit varied from 51.6 percent in 1938 to 20.8 percent in 1939. The range for Columnaris was from 29.5 percent in 1938 to 16.3 percent in 1937. The high proportion of flowers of the West Indian variety which developed into mature berries in 1938 was associated with a small crop of flowers.

This experiment was carried on in cooperation with the Agricultural Experiment Station of the University of Puerto Rico; Jaime Guiscafré Arrillaga, coffee specialist of the University Experiment Station, was in charge of the harvesting of this experiment.

ESSENTIAL-OIL INVESTIGATIONS

European war emphasizes development of essential-oil production in Western Hemisphere.

Many essential oils play an important role in the economy of the United States, not only for uses in drugs but also in the manufacture of soaps, cosmetics, and perfumes. The southern Mediterranean region, northern Africa, East Indies, and islands of the Indian Ocean are important geographic areas which normally supply the United States with a large part of its essential oils, and at the present time the war is making difficult the procurement of some of these oils. Not only from the standpoint of the security of national economy but also from the standpoint of developing better markets among tropical neighbors of the Western Hemisphere republics, research on the development of some of these crops has assumed great importance.

STUDIES OF LEMON-GRASS PRODUCTION AND PROCESSING

Full sunlight was essential to maximum yields of lemon grass.

The intensity of sunlight in the Tropics the year around is 25 to 40 percent greater than the maxima recorded by photometers in latitudes such as Washington and New York in the summer months.

In order to determine the effect of sunlight intensity on the tonnage and yields of oil per acre of lemon grass (*Cymbopogon citratus* (DC.) Stapf) an experiment was undertaken in which yields of the grass in full sunlight were compared with the yields in two-thirds sunlight, one-half sunlight, and one-third sunlight. Lath shade houses, as illustrated in figure 7, were used to permit the different degrees of sun-



FIGURE 7.—Lath framework used over plats of lemon grass to provide reduced intensities of sunlight.

light to reach the crop. There were four replications of each treatment with the plats arranged in a Latin square for statistical analysis. The grass was planted according to commercial practices and fertilized and harvested uniformly. The results of each treatment are shown in table 12.

TABLE 12.—Yield of grass, oil, and citral from 3 harvests of lemon grass, planted May 16-18, 1939, under four intensities of sunlight.

FIRST HARVEST, OCTOBER 2, 1939, PLANTS 4.5 MONTHS OLD

Treatment	Grass			Oil			Citral		
	Total weight harvested ¹	Yield per acre	Moisture content	In grass	Specific gravity	Calculated amount per acre	In oil	In grass	Calculated amount per acre
	<i>Pounds</i>	<i>Pounds</i>	<i>Percent</i>	<i>Percent</i>		<i>Pounds</i>	<i>Percent</i>	<i>Percent</i>	<i>Pounds</i>
Full sunlight.....	1, 147.00	28, 003.00	76.10	0.317	0.873	88.76	61.66	0.196	54.89
$\frac{2}{3}$ sunlight.....	847.50	20, 690.00	79.99	.281	.876	58.14	71.70	.202	41.79
$\frac{1}{2}$ sunlight.....	640.00	15, 625.00	81.75	.270	.878	42.19	75.00	.203	31.72
$\frac{1}{3}$ sunlight.....	455.00	11, 198.00	87.51	.235	.879	26.10	80.01	.188	20.88

SECOND HARVEST, JANUARY 9, 1940, RATOONS 3 MONTHS OLD

Full sunlight.....	917.25	22, 393.80	75.67	0.368	0.873	82.41	70.00	0.258	57.78
$\frac{2}{3}$ sunlight.....	774.00	18, 896.48	88.69	.335	.879	63.30	70.00	.235	44.41
$\frac{1}{2}$ sunlight.....	587.25	14, 337.16	81.69	.320	.879	45.88	74.00	.237	33.98
$\frac{1}{3}$ sunlight.....	353.75	8, 636.47	79.40	.326	.877	28.15	80.00	.261	22.54

THIRD HARVEST, APRIL 4, 1940, RATOONS 3 MONTHS OLD

Full sunlight.....	346.50	8, 459.47	75.96	0.222	0.907	18.78	72.50	0.161	13.62
$\frac{2}{3}$ sunlight.....	247.50	6, 042.48	79.60	.245	.916	14.80	76.50	.187	11.29
$\frac{1}{2}$ sunlight.....	169.00	4, 125.98	81.77	.194	.937	8.00	72.50	.141	5.82
$\frac{1}{3}$ sunlight.....	106.00	2, 587.89	88.10	.215	.981	5.56	65.00	.141	3.65

¹ From 4 randomized plats, each 0.01024 acre in area, per treatment.

Shading increased citral content in oil but decreased percentage oil and grass tonnage.

It can be seen from table 12 that, contrary to the effect of sunlight on such crops as coffee, the more sunlight reaching the lemon grass the greater the tonnage per acre and the higher the oil content. The percentage of citral in the oil had a tendency to improve with shading; however, because of the lower percentage of oil in grass under shade the percentage of citral in grass was lowest with most shade. Also, because of the much greater tonnage of lemon grass per acre obtained in full sunlight, the yield of citral per acre was much greater with full sunlight. These conclusions were applicable not only to the first harvest but also to the harvests of the first- and second-ratoon crops.

Lengthening growth period increased yield of lemon grass.

In yield experiments with lemon grass at this experiment station it has usually been the practice to make four harvests a year, one about every 12 or 13 weeks. In order to observe the effect of prolonging the growth period, an experiment was started in which plats of grass were allowed to grow for 12, 15, 18, and 21 weeks, respectively, before being harvested. There were eight replicated plats for each treatment. Planting, fertilizing, and harvesting operations were identical for all treatments. The yields of grass, oil, and citral obtained from each treatment are shown in table 13. These data are expressed graphically in figure 8.

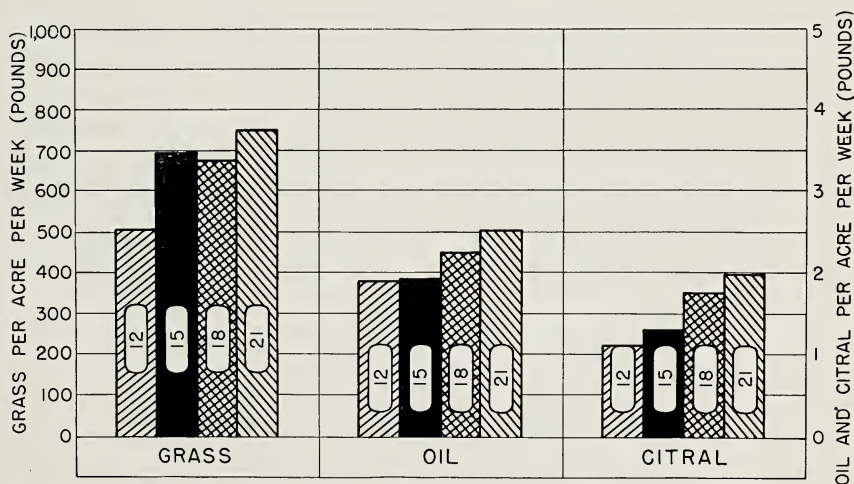


FIGURE 8.—Yields per acre per week of grass, oil, and citral from lemon grass harvested at different ages. Figures in bars indicate age at harvest in weeks.

TABLE 13.—*Yield of grass, oil, and citral from lemon grass planted Oct. 26, 1939, and harvested at different ages, 1940*

Date of harvest	Age at harvest	Yield of grass ¹ —		Oil—				Citral—			
		Per acre	Per acre per week of growth	In grass	Specific gravity	Calculated amount per acre	Calculated amount per acre per week of growth	In oil	In grass	Calculated amount per acre	Calculated amount per acre per week of growth
	Weeks	Pounds	Pounds	Percent		Pounds	Pounds	Percent	Percent	Pounds	Pounds
Jan. 18.	12	6, 113. 74	509. 48	0. 3690	0. 878	22. 56	1. 88	60. 00	0. 221	13. 51	1. 13
Feb. 8.	15	10, 385. 95	692. 40	. 2733	. 891	28. 38	1. 89	71. 00	. 194	20. 15	1. 34
Feb. 29.	18	12, 223. 28	679. 07	. 3361	. 892	41. 08	2. 28	76. 50	. 257	31. 41	1. 74
Mar. 21.	21	15, 916. 10	757. 91	. 3352	. 895	53. 35	2. 54	78. 50	. 263	41. 86	1. 99

¹ From 8 replicated plats, each 0.022383 acre in area, per treatment.

The longer the growth period the greater the yields of grass, oil, and citral per acre per week.

Table 13 shows that not only did the tonnage of lemon grass increase but also the trend of yields of lemon grass per acre per week definitely increased with longer growth periods. The percentage of oil in the grass was greatest with the shortest period of growth, but because of the much better yields of grass per acre per week the yield of oil per acre per week was also much better with the longest period of growth.

The percentage of citral in oil improved notably as the growth period of the grass was lengthened. This, together with the better yield of grass per acre per week, gave a much higher yield of citral per acre per week with the longest periods of growth.

The yields of oil per acre in all treatments in this experiment were unusually low as compared with acre yields in other experiments; this is attributed to the growth periods coinciding with the winter months of Puerto Rico, characterized by cooler weather and the dry season.

It is concluded from the experiment that it would be logical and most profitable to grow lemon grass, during the winter months at least, in the environment of Puerto Rico for periods of at least 21 weeks between planting or ratooning and harvesting.

A fertilizer-constituent experiment with lemon grass was conducted during the year.

An experiment to test the effect of various fertilizer constituents on tonnage and oil and citral contents of lemon grass was conducted during the year. Catalina clay, which is one of the most widely extended soil types in Puerto Rico, was selected for the experiment. There were six replications of the plats of each treatment in a Latin-square arrangement for statistical analysis.

All fertilizer constituents were applied at the rate of 100 pounds per acre; the nitrogen used was in the form of ammonium sulfate, phosphoric acid in the form of calcium superphosphate, and potash in the form of potassium sulfate. The lime was applied in the form of calcium hydrate. Fertilizers were applied at the bottom of the furrows just previous to planting, which was made on June 30, 1939. No additional fertilizer was applied for the ratoon crop. The first harvest was made January 4, when the plants were approximately 6 months old, and the second harvest on April 24, 3½ months later. The yields of grass, oil, and citral from each treatment are shown in table 14.

TABLE 14.—*Yields of grass, oil, and citral from the first and second harvests of lemon grass from plats receiving specified fertilizer treatments at planting on June 30, 1939*

FIRST HARVEST, JAN. 4, 1940, PLANTS 6 MONTHS OLD

Treatment	Grass		Oil			Citral		
	Total weight harvested ¹	Yield per acre	In grass	Specific gravity	Calculated amount per acre	In oil	In grass	Calculated amount per acre
	<i>Pounds</i>	<i>Pounds</i>	<i>Percent</i>		<i>Pounds</i>	<i>Percent</i>	<i>Percent</i>	<i>Pounds</i>
Check	1,300.00	15,318.62	0.412	0.874	63.11	80.00	0.330	50.55
N	1,456.25	17,159.80	.427	.871	73.27	73.00	.312	53.54
NP	1,573.25	18,538.48	.486	.870	90.10	78.00	.379	70.26
NK	1,959.00	23,083.99	.457	.873	105.49	76.00	.347	80.10
NPK	2,032.00	23,944.19	.500	.871	119.72	62.00	.310	74.23
NCaO	1,545.25	18,208.54	.427	.871	77.75	76.00	.325	59.18

SECOND HARVEST, APR. 24, 1940, RATOONS 3.5 MONTHS OLD

Check	435.50	5,127.30	0.303	0.8960	15.54	61.00	0.185	9.49
N	476.00	5,608.97	.230	.8880	12.90	65.00	.150	8.41
NP	485.00	5,715.03	.240	.8497	13.72	65.50	.157	8.96
NK	689.00	8,118.87	.265	.8914	21.52	66.50	.176	14.29
NPK	586.00	6,095.17	.288	.8828	19.89	70.50	.203	14.02
NCaO	451.00	5,314.39	.269	.9045	14.30	61.50	.165	8.77

¹ From 6 randomized plats, each 0.014144 acre in area, per treatment.

Potash gave increased tonnage of lemon grass on Catalina clay soils.

It is evident from table 14 that nitrogen alone considerably increased lemon-grass tonnage as compared with the unfertilized plats. Nitrogen plus phosphoric acid gave an appreciably greater yield but nitrogen plus potash gave a markedly improved yield. The complete fertilizer containing nitrogen, phosphoric acid, and potash gave the best yields of grass per acre in the plant crop, but the results were not consistent in the first ratoons.

The low yields in the ratoons as compared with the plant crop can logically be attributed to the 3.5-month growth period of the ratoons as compared with the 6-month growth period for the plant crop, and to the coincidence of the ratoons with the dry season in Puerto Rico, January to April, whereas the plant crop developed during the rainy season, June to December.

As regards yield of oil in the plant crop, nitrogen increased the percentages of oil in grass slightly, but nitrogen plus phosphoric acid gave an even greater improvement. Nitrogen plus potash materially increased the yield of oil, and the results with complete fertilizer substantiated the results with phosphoric acid and potash. It is notable that the yields of oil from the first ratoons were markedly lower than from the plant crop; this result, not obtained in previous experiments, was probably due to seasonal conditions existing during the present experiment.

In direct contradiction to the results with the plant crop, all fertilizer constituents lowered the percentages of oil in the ratoon crop as compared with the percentages from unfertilized grass. Of the fertilized plats in both plant and ratoon crops, the complete fertilizer gave the best yields of oil. All fertilizer constituents seemed to lower the specific gravity of the oil, but such differences were not

statistically significant. Nitrogen and phosphoric acid applications gave good increases per acre in yields of oil in the plant crop but decreases in the ratoon crop as compared with the check. Potash applications gave appreciable increases in yield of oil in both the plant and ratoon crops.

The percentage of citral in oil seemed to be inversely proportional to the percentage of oil in grass in this experiment. In the plant crop, where nitrogen, phosphoric acid, and potash increased the percentage of oil in grass, they simultaneously decreased the percentage of citral in the oil. In the first ratoons where these fertilizers applications decreased the percentage of oil in grass they increased the percentage of citral in the oil.

Nitrogen plus potash gave highest yields of citral per acre.

The percentage of citral in the grass was of course dependent upon the percentage of oil in the grass, and varying results were therefore obtained. The yields of citral per acre were dependent upon the percentage of citral in oil, percentage of oil in grass, and the yields of grass per acre. In the plant crop there were notable increases in citral per acre from applications of nitrogen, phosphoric acid, and potash, but because of the low percentage of citral in the oil in the plats receiving complete fertilizer the yield of citral per acre in such plats was correspondingly low. In the first ratoons the low percentage of oil in the grass gave lower yields of citral per acre from the applications of nitrogen and of nitrogen and phosphoric acid than the check plats. However, applications of nitrogen and potash gave the highest yield of citral per acre.

As a conclusion, it is indicated that applications of nitrogen and potash to lemon grass on Catalina clay soils resulted in statistically significant and profitable increases in yield of lemon grass, oil, and citral. The experiment also has significance for other crops on Catalina clay, inasmuch as it indicates a marked potash deficiency in this soil type.

None of the treatments used seemed to affect the solubility of the oil in alcohol, since samples from all plats possessed normal solubility.

As regards the results of applications of lime to lemon grass on Catalina clay, the conclusions are somewhat surprising, inasmuch as only 100 pounds of calcium oxide were applied per acre, a small treatment as compared with usual commercial applications. This small amount gave a significant increase in tonnage per acre of lemon grass and increased the percentage of oil in the plant crop, but caused decreases in the ratoons, as was the case with the fertilizer applications. The percentage of citral in the oil of the plant crop decreased following the application of lime, although not significantly so. The yield of citral per acre was increased appreciably in the plant crop but not in the ratoon crop by this small lime application.

Increased applications of nitrogen increased lemon-grass tonnage per acre.

In December 1939 a fertilizer experiment was initiated to test the effects of increasing quantities of nitrogen on yields of lemon grass, oil, and citral per acre. The nitrogen was applied in the form of ammonium sulfate and there were 10 replications of each treatment so

arranged as to provide for statistical analyses. As usual, the fertilizer was applied in the furrows prior to planting. The harvest results are given in table 15.

TABLE 15.—Yield of grass, oil, and citral from lemon grass harvested after 6 months from plats which received different amounts of nitrogen per acre at planting on Dec. 4 and 5, 1939

Nitrogen per acre (pounds)	Grass		Oil			Citral		
	Total weight harvested	Yield per acre	In grass	Specific gravity	Calculated amount per acre	In oil	In grass	Calculated amount per acre
	Pounds	Pounds	Percent		Pounds	Percent	Percent	Pounds
0	2,816.5	29,210.74	0.3616	0.8826	105.63	71.20	0.257	75.07
60	2,895.5	30,030.08	.3370	.8758	101.20	76.17	.257	77.18
120	2,958.0	30,678.28	.3481	.8797	106.79	74.66	.260	79.76
180	3,043.5	31,565.03	.3829	.8818	120.86	73.00	.280	88.38
240	3,180.0	32,980.71	.3684	.8915	121.50	78.00	.287	94.65

¹ From 10 randomized plats, each 0.009642 acre in area, per treatment.

Lemon grass responded to increased applications of nitrogen.

Table 15 shows that the lemon-grass tonnage per acre increased with increasing applications of nitrogen; the results were statistically significant. Where but 60 and 120 pounds of nitrogen per acre were applied the percentage of oil in the grass decreased as compared with the control plats. However, there was a significant increase in yield of oil from an application of 180 pounds of nitrogen per acre but not with 240 pounds. There were no significant differences between treatments as to the specific gravity of the oil obtained.

Citral yields per acre increased consistently with increased nitrogen fertilizer applications.

In the case of the percentage of citral in the oil, there were increases with the increasing nitrogen applications per acre, but these increases were not consistent. It is notable that the percentage of citral in the grass increased consistently with increasing applications of nitrogen per acre. The yields of citral per acre increased consistently as the applications of nitrogen per acre were increased, and the results were statistically significant. At present prices for ammonium sulfate the increases of lemon grass, oil, and citral were not profitable. However, the soils upon which these results were obtained were comparatively fertile; it is probable that on soils of lower natural fertility similar applications of nitrogen fertilizers would be profitable.

Sodium chloride increased the yield of oil obtained in distillation of lemon grass.

In the extraction of bay oil in the Virgin Islands sea water is sometimes used to generate the steam for the distillation of the bay-oil leaves. Where sea water is not available it is sometimes the practice to add salt to fresh water from which the steam is generated to pass through the bay-oil leaves. Preliminary trial runs showed that the addition of salt to the fresh water used for the generation of steam in the distillation of lemon grass gave greater yields and better qualities of oil than the use of fresh water alone. A preliminary experiment was therefore

carried out to measure the effect of the addition of different concentrations of sodium chloride to the water for the generation of steam in the distillation of lemon grass; in this experiment what has been called direct distillation was used, that is, the grass was immersed in the various concentrations of salt water during the distillation. The eight salt concentrations used varied from zero to 6 normal. The salt used was a locally available product recovered from sea water.

The grass was chopped into $\frac{1}{4}$ -inch pieces and thoroughly homogenized before sampling. Five samples of grass were distilled in each concentration of salt water. The yield and the results of the analysis of the oils obtained in these distillations are given in table 16 and presented graphically in figure 9.

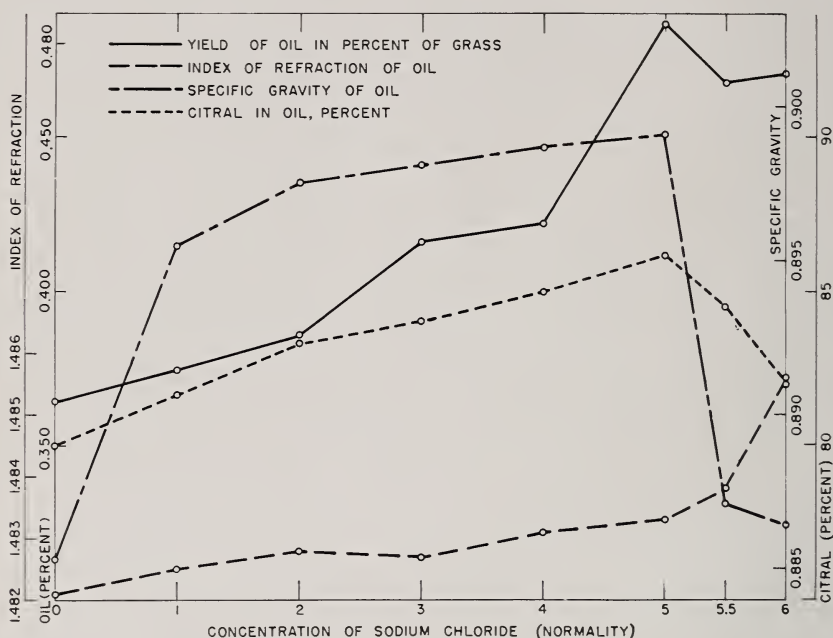


FIGURE 9.—Yield and analysis of lemon-grass oil obtained by distillation of the grass in different concentrations of sodium chloride in water.

TABLE 16.—Yield and analysis of lemon-grass oil obtained by distillation of the grass in water containing different concentrations of sodium chloride

Concentration of salt in distillation water (normality)	Amount of salt dissolved in 6 liters of water	Yield of oil ¹	Oil in grass	Analysis of oil ¹		
				Index of refraction	Specific gravity	Citral content
	Grams	Grams	Percent			Percent
0	0	33.0	0.364	1.4822	0.8853	80.00
1	351	34.0	.375	1.4825	.8955	81.66
2	702	35.0	.386	1.4828	.8975	83.33
3	1,052	37.7	.415	1.4827	.8981	84.00
4	1,403	38.3	.422	1.4831	.8987	85.00
5	1,754	39.6	.487	1.4833	.8991	86.20
5.5	1,931	42.5	.468	1.4838	.8871	84.47
6	2,106	42.6	.471	1.4855	.8864	81.99

¹ Average of 5 replicates.

It is evident in table 16 that the yield of oil increased as the substrata for distillation approached saturation with respect to the salt. The quality of the oil as measured by citral content and density also increased up to 5 normal salt solution and then dropped. The refractive index of the oil rose consistently as the solution approached saturation. It is apparent that when normality was above 5, constituents other than citral were contributing to the increase in yield and refractive index of the oil. The amount of citral produced was greatest at the salt concentration of 5.5 normal. In practice, the choice between recovery at 5 normal and 5.5 normal would depend on the economic aspects of production. Recovery at 6 normal resulted in an insignificant increase in yield of oil and a decrease in the amount of citral recovered.

Lemon-grass residue is a possible cattle feed.

In the foregoing distillation experiment, only sodium chloride was tried because it was inexpensive, and it was planned to feed the grass residue to dairy cattle. In a feeding experiment conducted by Antonio R. Villamil and Armando Arroyo, the Guernsey cows of the experiment station herd fed on this grass without hesitation, whereas the cows ate little lemon-grass residue when distilled without salt. The more complete extraction of the oil with salt, as well as the presence of the salt itself, made the residue more palatable; there was no evident effect on the milk flavor following the feeding of the lemon-grass pulp.

In the agronomic experiments with lemon grass in the foregoing, single representative 20-pound samples of grass from each plat were distilled. In the nitrogen-quantity experiment duplicate samples of the oil from each plat were analyzed; in the remainder of the experiments analyses were made of duplicate samples of the homogenized oil from similarly treated plats.

BAY OIL

Rock salt in distillation water increased yield and quality of bay oil.

One of the few essential oils which at the present time is produced commercially in Puerto Rico is that distilled from the leaves of bay (*Pimenta racemosa* (Nill.) Moore). A grove of young bay trees on the Las Mesas property of the experiment station is shown in figure 10.

In a preliminary experiment a quantity of 9-month-old leaves of bay grown at Patillas and testing free of lemon scent was used to compare the effect of three methods of distillation on the yield and characteristics of the resulting oils. After the usual preparation and homogenization, the leaves were weighed into samples of 8 pounds each, and duplicate samples were distilled by each method with the following results:

By the first method, which consisted in distilling the leaves immersed in water, i. e., direct distillation, an oil yield of 0.958 percent was obtained. In the second or ordinary steam-distillation method, in which steam was run through the leaves, the average oil yield was 1.011 percent. The third method was a modification of the first but, as in the experimental distillation of lemon-grass oil previously

described, the water in which the leaves were immersed contained 25 percent of rock salt by weight. By this modification 1.160 percent of oil was obtained from the bay leaves.

While a slightly higher yield of oil was obtained by steam distillation than by direct distillation, the highest yield resulted when the leaves were distilled in water containing salt. This is an important practical saving, since direct distillation is cheaper and simpler than steam distillation.



FIGURE 10.—Experimental plantation of bay trees on the Las Mesas property of the experiment station. Planted in 1938.

Bay oil obtained by distillation from salt solution possessed good chemical and physical properties.

The results of analyses of samples of the oil obtained by the three methods described above, and for comparison, analyses of standard commercial bay oil, are shown in table 17.

TABLE 17.—*Analysis of bay oils from different methods of distillation compared with the usual commercial standard*¹

Sample No.	Description	Yield of oil	Specific gravity at 15° C.	Index of refraction at 27° C.	Phenol	Color
		Percent			Percent	
1	Oil obtained by direct distillation	0.958	0.9910	1.5122	63.33	Light reddish yellow.
2	Oil obtained by steam distillation.	1.011	.9906	1.5122	65.00	Very light reddish yellow.
3	Oil obtained by direct distillation plus rock salt.	1.160	.9889	1.5131	70.00	Extremely light reddish yellow.
4	Standard commercial oil ²		.965 to .985	1.5100 to 1.5200	—	Light reddish yellow.

¹ All samples were soluble in equal volumes of 90-percent alcohol.

² From Parry (8, p. 387).

Table 17 shows that the oil obtained from the salt distillation had the lowest specific gravity, the highest phenol content, and the most desirable appearance of the three oils analyzed.

ILANG-ILANG

Ilang-ilang trees pruned monthly gave the highest yield of flowers.

In the report for 1939 an experiment was described which had as its purpose the reduction of harvesting costs of ilang-ilang (*Cananga odorata* Hook. f. and Thoms.) by pruning to secure low-headed trees. In the experiment the controls consisted of unpruned trees, a second lot consisted of trees pruned back monthly to form low-headed trees (from 8 to 9 feet in height), and the third lot consisted of trees pruned back annually to secure the same effect. During the past year it has been possible to obtain another series of harvest results. These are presented in table 18 and shown graphically in figure 11.

TABLE 18.—Weight of flowers from ilang-ilang trees under different pruning treatments

Sym- bol	Treatment	Flowers harvested ¹			Weight of flowers per acre from third harvest	Total weight of flowers per acre from the three harvests
		First har- vest, 1937-38	Second har- vest, 1938-39	Third har- vest, 1939-40		
		<i>Kilograms</i>	<i>Kilograms</i>	<i>Kilograms</i>	<i>Kilograms</i>	<i>Kilograms</i>
X	None.....	0.9077	13.799	20.4726	73.397	126.121
Y	Topped monthly.....	.2148	10.239	25.4508	91.244	128.724
Z	Topped yearly.....	.2958	7.502	12.4121	44.499	72.455

¹ From 6 plats of 9 trees each in each treatment, or 0.27893 acre.

It is evident from table 18 that in the third harvest the trees topped monthly gave a significantly greater yield of flowers. When all the results were compared and calculated on an acre basis, the treatments in which the trees were topped monthly gave the best yield of flowers per acre. The trees that were topped yearly have given the poorest yields of flowers in each crop since the inception of the experiment.

Hence from the foregoing results it appears that not only are labor costs greatly lowered by topping the trees monthly to minimize efforts in harvesting flowers, but also better yields per acre are obtained.

Agronomic investigations of essential-oil crops have been conducted by Antonio R. Villamil, junior agronomist; processing studies have been conducted by Noemí G. Arrillaga, junior chemist. Appreciation is expressed to Merriam A. Jones, junior chemist, and José O. Carrero, assistant chemist, for their cooperation in the foregoing studies.

BAMBOO PROPAGATION AND UTILIZATION

Multiplication of borer-resistant bamboo species was an essential activity.

The only clump bamboo now widely distributed in Puerto Rico is *Bambusa vulgaris* Schrad., the culms of which when cut are usually attacked by the powder-post beetle, *Dinoderus minutus* F. Although it is possible to reduce the susceptibility to this insect, it is not possible to secure complete resistance. It has therefore been the policy of the

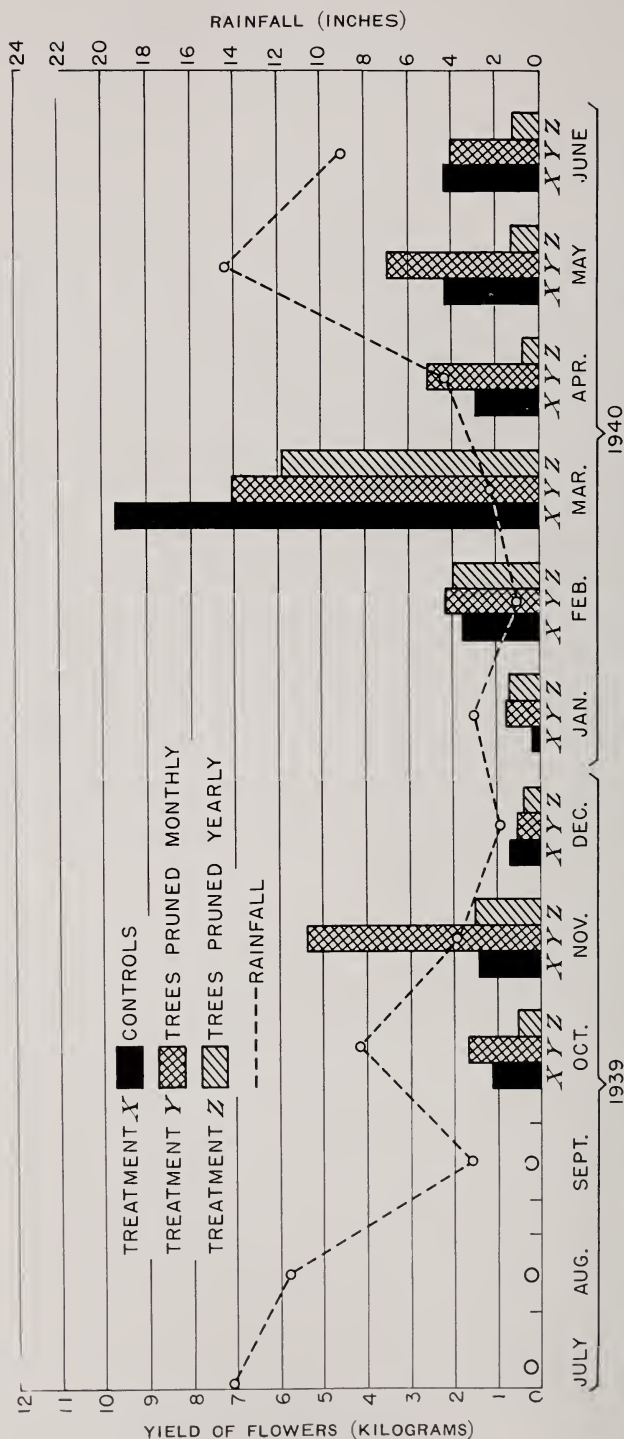


Figure 11.—Monthly yields of ilang-ilang flowers, during the third year, from trees under different pruning treatments, and corresponding rainfall. X, Y, and Z indicate treatments mentioned in table 18.

station not to use this species in construction, as if even a few articles became borer-damaged this might result in injuring the reputation of Puerto Rican bamboo products. The policy in this project, therefore, has been to propagate and multiply as rapidly as possible the newly introduced species of bamboo which have a high degree of resistance to the powder-post beetle. Such shop work as has been carried on has been mainly to maintain interest in the project and has been with little benefit of technical designing.

Methods of curing bamboos are now standard practice.

In the annual reports for 1936, 1937, and 1938 the results of experiments were recorded showing methods of minimizing borer attacks even in the more susceptible species. The procedures suggested as the results of these experiments have now been adopted at the experiment station as standard practice. All bamboos, whether susceptible or



FIGURE 12.—Irrigation reservoir at the experiment station, showing bamboo culms in process of submergence for 6 to 8 weeks to lessen their attractiveness to the powder-post beetle.

resistant species, when cut are allowed to stand upright in the clump with their leaves and branches uncut and intact for a period of 1 or 2 weeks. The side branches are then cut off and the bamboo poles are hauled to one of the station's several irrigation reservoirs where they are submerged in water for from 6 to 8 weeks. During this submergence there is a diffusion of the carbohydrates and other nutrients from the bamboo; often, also, the buds of the bamboo germinate and grow while in the water, thus further exhausting the nutrients from the wood and rendering it less attractive to the beetle.

On removal from the water the bamboos are allowed to dry and are placed in dry storage removed from further weathering effects. This practice has given good results even in most cases with the susceptible wood of our indigenous *Bambusa vulgaris*. Bamboo curing in a water reservoir is shown in figure 12.

New bamboo species were introduced during the year.

One plant of *Bambusa giganteus* Hort. was obtained from the New York Botanical Garden during the year. An additional plant of *Gigantochloa aspera* Hort. was obtained from the Division of Plant Exploration and Introduction of the Bureau of Plant Industry, and three plants of an undetermined bamboo species, Plant Introduction No. 126493, which originated in Calcutta, were obtained through the kindness of the same agency.

Plants of several bamboo species obtained from Dutch Guiana by Kenneth A. Bartlett of this station were housed on the plant quarantine greenhouse of the Division of Plant Exploration and Introduction in Washington awaiting shipment to Puerto Rico at the close of the fiscal year.

There are still a number of valuable industrial bamboos, not only in Asia and the East Indies, but also in parts of South America, which it would be desirable to obtain for multiplication for distribution throughout the Western Hemisphere.

Scale insects on bamboo were controlled by introduced insect predators.

The bamboo species indigenous to Puerto Rico, *B. vulgaris*, is unusually susceptible to the bamboo scale insects *Asterolecanium miliaris* (Bdv.) and *A. bambusae* (Bdv.). It is interesting to observe that at the close of the fiscal year under review these insects were moderately well under control at the experiment station due to the activities of the predators introduced from the continental United States, South America, and islands of the West Indies by K. A. Bartlett and H. K. Plank in 1936 and 1937. Of these predators, all coccinellids, *Egus platycephalus* Muls. from Cuba was most active, not only in distributing itself but also in preying on bamboo scales. *Curinus* species from Martinique and *Cladis nitidula* F. and *Pentilia castanea* Muls. from Trinidad were also notably active in the control of these pests.

Distribution of industrial bamboo species is now under way.

During the year, 239 small clumps of newly introduced bamboo species were distributed to such agencies as the Federal Forest Service, the Puerto Rico Reconstruction Administration, and the Agricultural Extension Service of the University of Puerto Rico. Nursery fields at the experiment station were in such condition at the end of the fiscal year that thousands of plants of the newly introduced, beetle-resistant, industrial species will be available for distribution in the next 12 months.

Appreciation is expressed to the Civilian Conservation Corps for personnel used in the field propagation and multiplication of the new bamboo species. Armando Arroyo, scientific aide, has been in charge of bamboo propagation and distribution during the year.

Construction of split-bamboo fishing rods has industrial possibilities for Puerto Rico.

Using split bamboo of the species *Bambusa tulda* Roxb., some excellent fishing rods, such as those shown in figure 13 were constructed in the bamboo shop during the year. These rods have been found to be satisfactory under rigorous conditions of deep-sea fishing.



FIGURE 13.—Split-bamboo fishing rods for deep-sea fishing constructed from the wood of *Bambusa tulda* at the experiment station.

There would appear to be possibilities for a small industry in the manufacture of such rods from our best industrial bamboo species. There is an import tariff of 45 percent ad valorem on articles manufactured of bamboo entering the United States, which would give

Puerto Rico an advantage over other tropical countries in this small industry. This development is in line with the policy of industrializing in Puerto Rico its agricultural products. There would appear to be some advantages also in the manufacture of these fishing rods at the place where the bamboo is produced and processed.

Other new articles manufactured of bamboo during the year were small coin banks made of individual nodes of bamboo, attractive and intriguing to small children. A new type of cigarette box for desk use, attractive and distinctive to the Tropics, was constructed.

Appreciation is expressed to the National Youth Administration for considerable help in the shop work on bamboo during the year. James K. Alvis, assistant agricultural engineer, and Pedro A. Folch, junior agricultural engineer, have been in charge of bamboo shop work.

INVESTIGATIONS OF INSECTICIDAL PLANTS

Rotenone insecticides fill urgent needs of United States agriculture.

During the past 15 years there has been an urgent need in the United States for an effective insecticide, harmless to humans and domestic animals, that could be substituted for the inorganic insecticides containing lead and arsenic which leave toxic residues on the crops to which they are applied. Rotenone-containing products, milled from the roots of tropical plants of the two genera *Lonchocarpus* and *Derris*, effectively fill such specifications for a number of insect pests.

During the past few years the quantity of rotenone products used as insecticides in the United States has materially increased; during 1939 the United States imported approximately 2,000,000 pounds of *Derris* root grown in southeastern Asia and the neighboring islands, and an estimated 2,700,000 pounds of *Lonchocarpus* root from South America, where it is harvested from uncultivated areas and is known as "cubé," "barbasco," and "timbó."

Extensive collections of both *Derris* species from the Eastern Hemisphere and *Lonchocarpus* species from the Western Hemisphere have been assembled at this experiment station for comparative production studies. Most of these thrive under certain Puerto Rican environmental conditions, and it is possible that for a period of years, with some research advantages, the industry may thrive here. There are no import tariffs on these products entering the United States, so that with farm labor costing from 6 to 10 times more in Puerto Rico than in the East Indies, competition will be difficult without research advantages.

AGRONOMY OF ROTENONE CROPS

Trellising increased the yield of cuttings and roots of *Derris elliptica*.

Derris makes a vinelike type of growth, some of the branches attaining a length of as much as 50 feet under favorable conditions. The plants may either be allowed to trail along the surface of the soil, or they may be trained up on a trellis.

In June 1937 a replicated experiment with *Derris elliptica* (Roxb.) Benth. was initiated at the station to study the effect on yields of

trellising versus allowing the plants to trail along the ground. The trellises consisted of living support stakes of "bucare" (*Erythrina berteroana*), a tree of high vegetative vigor, to which were fastened three strands of galvanized-iron wire. Yields were measured by number of vine cuttings, weight of roots, and percentages of rotenone and total extractives in the roots. In September 1939, 27 months after planting, it was found that trellising had markedly increased the number of vine cuttings, the air-dry weight of roots, and the quantity of rotenone and total extractives contained in the roots. The concentration of rotenone and total extractives per unit of oven-dry weight of roots, however, was higher in roots of trailing plants than in the roots of those that were trellised.

Trellising decreased the concentration of rotenone and total extractives in the roots of Derris.

Training the plants on wire trellises 4½ feet high with the rows 4 feet apart resulted in the production of 304 percent more vine cuttings 9 inches in length than when the plants were not trellised. Trellising increased by 989 percent the number of cuttings with a diameter larger than 1.5 centimeters, and by 273 percent those with diameter between 1 centimeter and 1.5 centimeters. Trellising increased the air-dry weight of roots by 75 percent, the total quantity of rotenone produced by 52 percent, and the quantity of total extractives by 57 percent. However, the concentration of rotenone in the oven-dry roots was 14 percent, and of total extractives 11 percent higher for trailing plants than for those that were trellised. These differences in favor of the trailing plants were probably the result of the higher proportion of small roots they produced, for small roots usually contain higher percentages of rotenone and total extractives than large ones.

High water table reduced the concentration of toxic constituents in roots of Derris.

This experiment was planted on the heavy-clay lowland soil of the experiment station. Although surface drainage was good, during the periods of heavy rainfall the water table came to within 6.8 inches of the surface of the lowest plat, whereas in the highest plat the highest recorded level was 32.8 inches below the surface. Superficial blackening of many roots occurred in the three plats in which the water table was nearest the soil surface. The roots from these plats were lower in percentages of rotenone and total extractives than those from plats with lower water-table levels.

The yields obtained in this experiment are summarized in table 19.

TABLE 19.—Mean yield of cuttings, air-dry roots, rotenone, and total extractives per acre of trellised and trailing 27-month-old Derris elliptica plants

Product	Trellised plants	Trailing plants	Increase or decrease
			Percent
Small cuttings ¹number.....	113, 946±6, 544	30, 550±2, 846	273
Large cuttings ²do.....	15, 060±1, 530	1, 383± 322	989
Total cuttings.....do.....	129, 006±7, 530	31, 933±2, 933	304
Air-dry roots.....pounds.....	3, 040± 74	1, 738± 67	75
Rotenone ³percent.....	3.72	4.25	-11
Total extractives ³do.....	12.33	13.72	-11

¹ Between 1 and 1.5 cm. in diameter.

² Larger than 1.5 cm. in diameter.

³ Dry-matter basis, weighted average.

A general view of the experiment just before harvest is shown in figure 14, and the manner in which the roots were harvested is indicated in figure 15. Figure 16 shows the average yield of roots per $32\frac{1}{2}$ -foot row for trailing and for trellised plants.

Nine high rotenone-yielding clones of *Derris elliptica* were propagated.

By special arrangement with the Goodyear Rubber Plantation Company of Panama, the station during the year obtained propagating material of 9 superior clones of the Changi No. 3 variety of *Derris elliptica*, all of which originated in the Netherlands East Indies. A total of 287 plants of these 9 clones has been established, with the number of plants per clone varying from 29 to 41. The



FIGURE 14.—General view of the trellised-trailing experiment with *Derris elliptica* just before harvest. The five trellised plats and all but one of the trailing plats are shown.

parent stocks of 3 of the clones were reported to contain 10 to 11 percent of rotenone, slightly more than twice the percentage in the best stocks of *D. elliptica* in the station collection prior to the accession of these new clones. Plants of all 9 new clones are growing vigorously under Mayaguez conditions.

High rotenone-yielding clones of *Lonchocarpus* have been increased rapidly by budding.

During the summer of 1937 three plants of *Lonchocarpus* were found, the roots of which were higher in rotenone than is common for this plant; the roots of one plant contained 16.7, those of another 14.0, and those of the third 8.2 percent of rotenone. During the past $2\frac{1}{2}$ years these three plants have been handled as clones and increased as rapidly as possible by budding them onto established plants of the same and closely related species of lower rotenone content. When many of the scions resulting from these bud grafts had developed 40 or more usable buds, the buds on the scions were utilized for additional budding operations. At the time such a scion was removed

from the stock plant, one or two nodes were left above the graft union so that additional propagating material of the desired clone might be produced.



FIGURE 15.—A hillside plow drawn by oxen was used to loosen the roots in the soil. Owing to the compact texture of the soil it was necessary to use potato forks to complete the removal of the roots. In the left background two men are harvesting the last row of trellised plants of plat No. 5.

To date 351 successful bud grafts of these three clones have been made. At the end of June 1940 the scions developing from the successful bud grafts bore a total of 11,075 buds sufficiently well-developed for use in future budding operations, and 53 young scions had not yet produced mature buds. A summary of the results of these budding operations is given by clones in table 20.



FIGURE 16.—The average yield of roots per $32\frac{1}{2}$ -foot row of trailing and trellised treatments. The roots were sorted into two sizes—those 1 centimeter or less in diameter and those more than 1 centimeter in diameter. A and B were taken from trailing plants and C and D from trellised plants.

TABLE 20.—Increase in three superior clones of *Lonchocarpus* by bud grafting by June 30, 1940

Item	Clone A. 16.7 percent of rotenone	Clone B, 14.0 percent of rotenone	Clone C, 8.2 percent of rotenone	Total
	<i>Number</i>	<i>Number</i>	<i>Number</i>	<i>Number</i>
Successful bud grafts.....	66	283	2	351
Scions that produced usable buds.....	56	240	2	298
Young scions without usable buds.....	10	43	0	53
Total usable buds developed ¹	1,501	9,012	562	11,075

¹ Buds sufficiently mature June 30, 1940, to use in budding operations.

PHYSIOLOGY OF ROTENONE CROPS

Agronomic practice is based on physiology.

In the culture of any crop plant it is of primary agronomic importance to know the conditions favoring the elaboration of the compounds for which the crop is grown and to determine whether these compounds, once stored in the plant body, are at times changed by the plant into other substances of no economic value. In the 1939 annual report evidence was presented that the greatest elaboration of rotenoids ² is associated with flushes of growth and that, once stored in the plant body, rotenoids are metabolically inert. Investigations of the past year have further substantiated and clarified these views. The culture of rotenone crops based on these views might well be expected to diverge from that employed with crops grown for their carbohydrates, such as sugarcane.

Weather cycles influenced the tissue patterns of locally grown *Derris*.

Observations on the influence of weather cycles on the development of tissue patterns in the roots of *Derris* were continued. In general, there was a tendency for the production of a relatively high proportion of rotenoid cells to be associated with periods of rapid growth of the plants, such as is usually encountered in Puerto Rico in the spring soon after the beginning of the rainy season. During the dry season, when vegetative extension of the plants is at a minimum, comparatively few rotenoid cells were produced and cells filled with starch predominated in the xylem parenchyma formed during this period. On drying, the starch-filled tissues remain whitish whereas rotenoid cells turn dark. As a consequence, under conditions of alternately light and heavy rainfall in Puerto Rico, dried roots of *Derris* generally show a pattern of alternating bands of light and dark color that bears a close resemblance to that of growth rings in trees.

However, modifications of this tissue pattern, such as a wide band of rotenoid tissue being interspersed with narrow bands of starch tissue, are not infrequent. In such cases it seems likely that the modification might result from the periodic short droughts that frequently occur in Mayaguez during the rainy season. Roots that have unusually wide bands of rotenoid cells and comparatively little starch

² The term rotenoids, which has recently come into use, refers to rotenone and related compounds that react positively to the Durham test. Cells in which these compounds are stored are called "rotenoid cells" instead of "resin cells" as in previous reports. The terms "rotenoids" and "total extractives" are not synonymous as the total extractives include resinous and fatty substances that do not react positively to the Durham test.

tissue frequently do not fit completely into the picture of cyclic growth. Although no experimental data can be presented on the factors involved in variations of the latter type, it seems probable that internal conditions favoring an unusually extended period of rapid growth were operative in such cases, as, for example, the localized reutilization of the general store of food reserves.

Root diameter is not a reliable indicator of rotenoid content.

Variations of the last type of tissue pattern referred to in the preceding paragraph resulted in a marked increase in rotenoid content as demonstrated by chemical analysis. In one clone of *Derris elliptica*, roots with large numbers of rotenoid cells were found to contain twice the concentration of rotenoids as roots with few rotenoid cells. These extreme variations in composition were observed in roots of all sizes; frequently two roots of the same diameter on the same plant represented the two extremes of rotenoid- and starch-cell predominance.

It is apparent, then, that the relationship previously considered as existing between root diameter and rotenoid content needs modification. Until now it has been accepted that fine roots contain little rotenone, that as the roots enlarge their rotenoid content increases until they reach a diameter of 5 to 7 millimeters, and that then the rotenoid content declines as root thickening continues. It is only by the use of comparatively large samples that the effect of variations of rotenoid content among roots of the same diameter can be averaged and the chemical analyses give the so-called normal distribution of rotenoids.

Derris from the Far East showed tissue patterns similar to those of locally grown roots.

Examination of the weather records of Malaya indicated that similar tissue patterns should develop in the roots of *Derris* grown in that part of the world. Through the courtesy of the Cooperative Grange League Federation Soil Building Service, Inc., four lots of *D. elliptica* roots that originated in British Malaya and French Indochina were examined in their New York City warehouse. In these roots were found not only the main tissue patterns described above but also striking variations in the proportion of rotenoid and starch tissues in roots of the same diameter.

The lack of recognition of the wide variation that occurs in rotenoid content of roots of comparable diameters has doubtless been a critical factor in the failure of seller and buyer of *Derris* and *Lonchocarpus* roots to secure chemical analyses that agree. Representative tissue patterns of the roots of *Derris* grown in Puerto Rico and in the Far East are shown in figures 17 and 18, respectively.

Relation between carbohydrates and toxic constituents in *Derris* and *Tephrosia* roots was studied chemically.

Numerous local observations on sections of roots have supported the results of research in East Africa showing that starch and rotenoids are found in mutually exclusive cells in the roots of rotenone-producing plants. To what extent this relationship would be reflected in chemical analyses is shown in the following section of this report. The details of experimental procedure involved and the effects of

the treatments on the rotenone and total extractives content of the roots were given in the annual report for 1939; the present discussion is confined entirely to the relationship between carbohydrates and toxic constituents.

Method of sugar analysis was modified by addition of glucose.

The carbohydrate analyses were made using the Official and Tentative Methods of Analysis of the Association of Official Agricultural Chemists. Total sugars were determined by the Lane-Eynon volumetric method, using acid hydrolysis for disaccharides. Dextrins and starch were determined together, using malt diastase as the initial hydrolyzing agent. All mention of the carbohydrate component of roots of *Derris* in this report refers to total sugars, dextrins, and starch, considered as a unit.

In the Lane-Eynon method for the determination of sugars the sugar solution to be titrated must be within the concentration range of 1 to 8 grams per liter. The sugar content of most of the samples of *Derris* roots was so low that it was necessary to modify the method of analysis by adding pure glucose to the prepared solutions to bring



FIGURE 17.—Variation in the pattern of starch and rotenone cells in the roots of 2-year-old plants of the St. Croix variety of *Derris elliptica* grown in a sandy field on the estate of Clara E. Livingston near Dorado, P. R. Note the regularity with which all roots during the first year formed a central core of tissue high in rotenone and that this core was surrounded by tissue relatively high in starch. The pattern of second-year tissue varied as described in the text.

them within the specified range and subsequently subtracting the added glucose from the result. By this procedure all error of titration is reflected in the data of original sugar content. As a consequence, a minimum amount of sugar must be present in the original sample to keep the error of titration at a low value.

In order to determine this minimum, 10 titrations of a solution containing 1.023 grams of glucose per liter were made. On the basis of a standard deviation of 0.001 gram per liter the odds were approximately 19 to 1 that any titration of the same solution would not fall outside the range of 1.023 ± 0.002 grams per liter. On this basis, in order to restrict the relative error to less than 1 percent, the concentration of invert sugar in the sample must be at least 0.2 gram per liter before glucose is added.

This modified procedure was tested on a derris-root extract containing about 2 percent invert sugar. Without glucose added, the extract was found to contain 1.985 ± 0.0002 percent of invert sugar, and with glucose, 2.004 ± 0.002 percent after allowance was made for

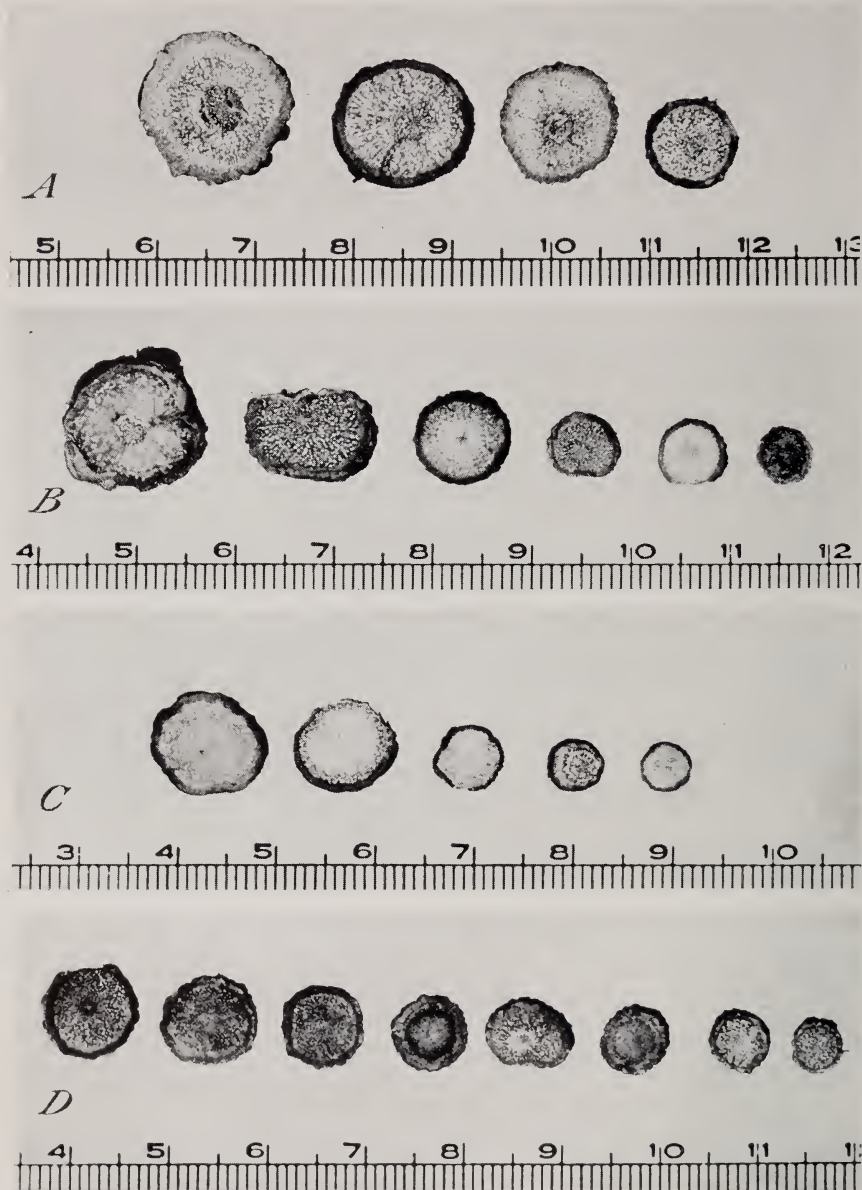


FIGURE 18.—Variation in pattern of rotenoid and starch cells in roots of *Derris elliptica* received from the Far East. A, Sacked roots from French Indo-China reported to contain 5.1 percent of rotenone. B, Baled roots from British Malaya reported to contain 5.7 percent of rotenone and 14.9 percent of total extractives. C, Baled roots from British Malaya reported to contain 7.8 percent of rotenone. D, Baled roots of uncertain specific origin in the Far East reported to contain 13 percent of rotenone and 30.6 percent of total extractives. Note especially the degree of discoloration of this lot of roots in comparison with that of the other three lots, showing that the extent of discoloration is an indication of rotenone and total extractives content. It is possible that none of these lots of baled or sacked roots represent either clones or pure varieties. Nevertheless, the similarity of tissue pattern in these lots to those of known purity is definite.

glucose added. Although the difference between the means of the two methods is statistically significant, it is of no appreciable consequence from the chemical standpoint inasmuch as the difference amounts to only approximately 1 percent.

In normal plants of *Derris elliptica* carbohydrates and toxic constituents were inversely correlated.

In the first experiment in which carbohydrates were studied the roots from 10 plants of a clone of the St. Croix variety of *Derris elliptica* were sorted into different size groups on the basis of their diameters, and their reaction to the Durham test determined. In Croix variety of *Derris* the concentration of carbohydrates tends to those that were only moderately positive, were analyzed for rotenone, total extractives, and carbohydrates. The results of the analyses are shown on a dry-matter basis in table 21.

TABLE 21.—*Rotenone, total extractives, and carbohydrates on a dry-matter basis in roots of corresponding diameters differing in reaction to the Durham test*

[All roots were selected from 1 set of 10 plants of *Derris elliptica* variety St. Croix]

Size group	Range of diameters	Rotenone		Total extractives		Carbohydrates ¹	
		Strongly positive to Durham test	Moderately positive to Durham test	Strongly positive to Durham test	Moderately positive to Durham test	Strongly positive to Durham test	Moderately positive to Durham test
	Mm.	Percent	Percent	Percent	Percent	Percent	Percent
Fine.....	1.9 or less.....	4.6	2.4	21.3	12.3	(²)	(²)
Small.....	2.0 to 4.9.....	6.3	2.8	25.2	13.8	9.6	14.7
Medium.....	5.0 to 9.9.....	6.3	2.6	24.6	11.1	13.7	18.1
Large.....	10.0 to 19.9.....	3.7	1.9	15.5	8.4	17.0	18.1
Very large.....	20.0 to 24.9.....	2.0	1.2	12.8	7.9	18.3	20.0
Largest.....	25.0 or more.....	1.6	1.8	8.1	7.7	16.1	16.4
All roots, average.....	3.8	2.1	16.3	10.0	³ 16.0	³ 17.8

¹ Carbohydrates include total sugars, dextrins, and starch.

² No fine-root material was available for carbohydrate analysis.

³ The weight of the fine-root fraction was not included in calculating these percentages.

It can be seen from table 21 that for any one size group the concentration of rotenone and total extractives was greater and that of carbohydrates was less in the roots that were strongly positive to the Durham test than in those that were only moderately positive. The difference in carbohydrates was proportionately less than that for the other two constituents; this was particularly true for roots of about 15 millimeters in diameter and larger. The results of this experiment are shown graphically in figure 19.

The inverse correlation between the concentration of carbohydrates and that of rotenone and total extractives in roots between 2 and 25 millimeters in diameter is readily apparent from figure 19. The results of these experiments are evidence that in normal plants of the St. Croix variety of *Derris* the concentration of carbohydrates tends to be inversely correlated with the concentration of rotenone and total extractives.

Differences in growth did not materially affect reciprocal relation of carbohydrates to rotenone.

In a second experiment the roots of 10 normal plants in each of two adjacent trellised rows of *Derris elliptica* var. Changi No. 3 were harvested separately. Chemical analyses of the several root fractions

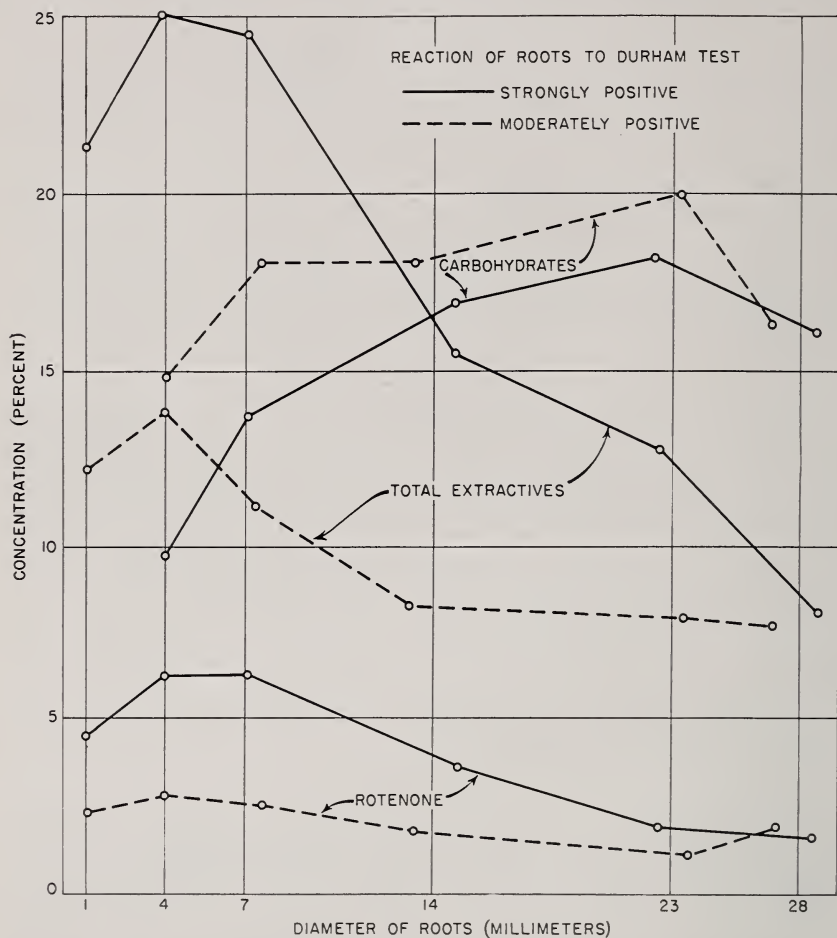


FIGURE 19.—Percentages of carbohydrates, total extractives, and rotenone on a dry-matter basis in roots of the St. Croix variety of *Derris elliptica* of corresponding diameters differing in reaction to the Durham test. All roots were selected from 1 set of 10 plants. The points on the graph represent the mean diameters for the roots in each size group. The mean diameters plotted for the fine-root fractions were estimated.

are given in table 22. Row 1 was the south-facing border row of the trellised area where less competition favored greater vegetative growth than occurred in row 2. As a result of its positional advantage, row 1 produced 37 percent more roots than row 2. With this increased production of roots were associated an increase in the average percentages of rotenone and total extractives and a decrease in the average percentage of carbohydrates. Thus, the general reciprocal relation

of rotenone and total extractives to carbohydrates was maintained in the averages for all roots of rows 1 and 2. However, some deviation from this relationship appeared when roots within individual size groups in the two rows were directly compared.

TABLE 22.—*Rotenone, total extractives, and carbohydrates on a dry-matter basis in roots of corresponding diameters from 10 plants in each of 2 adjoining rows of Derris elliptica var. Changi No. 3*

Size group	Range of diameters	Rotenone		Total extractives		Carbohydrates ¹	
		Row 1	Row 2	Row 1	Row 2	Row 1	Row 2
	Mm.	Per-cent	Per-cent	Per-cent	Per-cent	Per-cent	Per-cent
Fine.....	1.9 or less.....	4.5	5.0	11.8	12.6	2.6	24.0
Small.....	2.0 to 4.9.....	6.4	5.5	15.4	13.3	9.9	30.1
Medium.....	5.0 to 9.9.....	5.3	5.3	12.7	13.0	16.1	33.5
Large.....	10.0 or more.....	5.0	4.1	11.9	10.1	15.9	32.8
All roots, average.....		5.2	4.7	12.5	11.7	14.1	31.5

¹ Carbohydrates include total sugars, dextrins, and starch.

INVESTIGATIONS OF OTHER INSECTICIDAL PLANTS

Five introductions were added to the collection of insecticidal plants.

Seeds of four introductions of *Tephrosia vogelii* Hook. f.—Plant Introduction Nos. 125546, 126547, 126548, and 128996—were received from the Division of Plant Exploration and Introduction of the Bureau of Plant Industry and a small supply of seed of *Mundulea sericea* (Willd.) A. Chev., the East African fish-poison plant that produces rotenone in both roots and stems, from the Plant Culture League of San Pedro, Calif. These and three additional species of fish-poison plants—*Tephrosia candida* DC. (P. I. No. 118676), *T. vogelii* (P. I. Nos. 119087 and 126550), and *Tephrosia adunca* Benth. (P. I. No. 117931)—were propagated from seed during the fiscal year, producing 527 seedlings. All of these introductions are among those of which plant material has not yet been submitted to the cooperating bureaus of the United States Department of Agriculture for routine toxicological tests.

Herbarium specimens were deposited in Washington.

Herbarium specimens of 69 introductions of fish-poison plants were sent for identification to the Division of Plant Exploration and Introduction, Bureau of Plant Industry. E. P. Killip of the United States National Museum identified the species of *Derris* and *Lonchocarpus* included in this shipment; all other specimens were identified by taxonomists in the Division of Plant Exploration and Introduction.

Roots, stems, and leaves of a South American fish-poison plant, *Dahlstedtia pinnata* Malme, collected by K. A. Bartlett on a trip to Brazil, were completely negative to the Durham tests for rotenone and certain related compounds.

Rufus H. Moore, associate plant physiologist, and Merriam A. Jones, junior chemist, were in charge of investigations of insecticidal plants during the year. M. S. Lowman, Division of Drug and Related

Plants, Bureau of Plant Industry, made rotenone and total extractives determinations for all but the trellis-trailing experiment.

VEGETABLE CROP INVESTIGATIONS

There are three serious insect pests of sweet corn ears in Puerto Rico.

Varieties of sweet corn developed for production in the continental United States do not grow well in Puerto Rico and rarely produce any appreciable quantity of marketable ears. As a result of the activities of this experiment station U. S. D. A.-34 sweet corn, a variety that grows vigorously and yields heavily in the Tropics, was developed and seed was made available to island farmers in 1934. For a period of 6 to 7 months each year, during the late fall, winter, and early spring months, there is no fresh green sweet corn available on the New York market and the markets of the other large cities of the continental United States. However, Puerto Rican farmers have made little effort to supply these markets with sweet corn, largely because of the heavy infestations of the ears by three insect pests, the corn earworm (*Heliothis armigera* Hbn.), the fall armyworm (*Laphygma frugiperda* (S. & A.)), and the corn-silk fly (*Euxesta stigmatias* Loew).

Effective method of controlling corn ear pests was developed.

During the fiscal year 1939 a series of experiments was conducted in an attempt to develop an effective and economical control of these three pests. An account of the results of these experiments is given in the annual report of the station for 1939. Additional experiments were conducted along this same line during the past fiscal year.

The results of the 1940 experiments have confirmed those of the previous year in indicating that satisfactory control of these corn ear pests can be effected by the application, at the time the silks begin to wilt noticeably, of a 1:5 mixture of alcoholic pyrethrum extract in light, highly refined mineral oil to the silks of the ears at the point where they enter the husks. The experiments have demonstrated further that derris extracts could not be advantageously substituted either in whole or in part for the pyrethrum extract in the insecticidal mixture, and that water could not be used as a substitute for the mineral oil in the pyrethrum-extract-mineral-oil mixture. The details of the results of these and of previous experiments have been published in a circular of the station (2). The application of the results of these experiments should go far toward making it possible for Puerto Rican farmers to produce sweet corn for marketing in the continental United States during the winter months, inasmuch as the control of these ear pests was the last major problem confronting the farmers in the production of this crop.

The use of U. S. D. A.-34 sweet corn has continued to spread.

The use of U. S. D. A.-34 sweet corn has continued to spread throughout the island. During the past year the Agricultural Extension Service of the University of Puerto Rico and the Vocational Agriculture Service of the Insular Department of Education have taken an active interest in this promising new crop for Puerto Rico. As a consequence, considerable impetus has been given to the dis-

semination of information about the crop, and increased plantings have resulted. Until this past year the experiment station has been the only source of U. S. D. A.-34 sweet corn seed in Puerto Rico. During the year, as a result of seed made available by the station, both the Extension Service and Vocational Agriculture have produced liberal quantities of seed for distribution to the farmers of the island. Consequently it is anticipated that in the future more emphasis can be devoted to the improvement of the variety at the experiment station and less to the production of seed for distribution.

Attempts to produce seed of continental American varieties of corn in Puerto Rico during winter months have failed.

During recent years producers of seed of highly developed hybrid field corn in the continental United States have shown increasing interest in speeding up their breeding operations by locating a place where corn seed can be increased advantageously during the winter months. During the 1939 fiscal year the experiment station, in co-operation with Frederick D. Richey, of Ashville, Ohio, began an investigation of the possibilities of the utilization of Puerto Rico as a winter seed-production center for this crop. This cooperative undertaking was continued during the past year. For the second year in succession unsatisfactory seed yields were obtained. The results with continental American varieties of corn during the past 2 years are in keeping with the results of previous less extensive trials at the station and elsewhere on the island.

At present it seems unlikely that strains and varieties of corn developed for use on the continent can be grown successfully in Puerto Rico during the winter months unless a satisfactory method of controlling *Helminthosporium* leaf spot can be worked out.

Cantaloups of high edible quality were produced.

For the second year in succession, acceptable yields of high-quality cantaloups have been obtained from plantings made on the experiment station grounds during the winter months. The varieties grown during the past year were Hale's Best, Cooper's Sweetheart, Green-Fleshed Rocky Dew, and Orange-Fleshed Rocky Dew. There are no cantaloups on the markets of the continental United States for a period of approximately 6 months each year, during the late fall, winter, and early spring. The possibilities of the commercial production of cantaloups in Puerto Rico during the winter months are being investigated more in detail by the station. The results with this crop to date have been encouraging and, unless some unforeseen limiting factor in its production should appear, it shows promise of becoming a valuable new winter vegetable crop for export to the continental United States.

Wallace K. Bailey, horticulturist, on leave during a large part of the year, has directed the plantings of vegetable crops by correspondence.

SUGARCANE INVESTIGATIONS

Sugarcane research activities have been reduced.

As in the past few years, it has not been possible to give much attention to sugarcane investigations, it being the policy of the station to devote its resources as much as possible to developing new crops

for island economy. A few cooperative field agronomic tests are being carried on, however, one of which was harvested, and the results of which follow:

First-ratoon crop in sugarcane-variety trial on Toa silty clay at Añasco was harvested in March.

In the annual report for 1939 were recorded the results of the plant crop harvested in a field trial of sugarcane varieties that had been set out on Toa silty clay at Añasco on August 11, 1937. This experiment was conducted by E. H. Barrow, superintendent of the Añasco district of Russell & Co., and, as in 1939, in cooperation with this station. One Fajardo Central variety, F. C. 916, and three Mayaguez varieties—Mayaguez 270, Mayaguez 275, and Mayaguez 317—were tested against the district standard, P. O. J. 2878. Each variety was replicated five times in $\frac{1}{20}$ -acre plats arranged in a Latin square for statistical analysis.

Following the harvest of the plant crop on February 28, 1939, ammonium sulfate was applied to all plats at the rate of 400 pounds per acre March 30, and at 200 pounds per acre April 27. The first-ratoon crop was harvested March 27, 1940, at 13 months. Each plat was cut and weighed separately, but cane from the five replicated plats of each variety was milled as one lot, and the juice analyses were made of samples from this composite milling.

Mayaguez 275 outyielded other varieties in first-ratoon crop.

It is evident from table 23 that in the first-ratoon crop Mayaguez 275 produced more sugarcane per acre than any other variety; this superiority was highly significant. Another striking comparison brought out in the test was the reversal of the cane-tonnage superiority held by Mayaguez 270 in the plant crop. In that crop Mayaguez 270 outyielded all the other varieties, but in the first-ratoon crop it was outyielded by all the other varieties. Furthermore, in the first-ratoon crop Mayaguez 270 was the only variety that produced less cane than the standard P. O. J. 2878.

TABLE 23.—*Summary of average yields of cane and sugar per acre and sucrose in juice from plant and first-ratoon crops of varieties tested on Toa silty clay of Russell & Co., Añasco, 1939 and 1940*

Variety	Plant cane, 18½-month crop ¹			First ratoon, 13-month crop ²			Totals for two crops		Average per acre per month	
	Cane per acre	Sucrose in juice	Sugar per acre	Cane per acre	Sucrose in juice	Sugar per acre	Cane per acre	Sugar per acre	Cane	Sugar
	Tons	Percent	Tons	Tons	Percent	Tons	Tons	Tons	Tons	Tons
F. C. 916.....	58.12	14.3	6.28	44.71	16.0	5.43	102.83	11.71	3.26	0.372
Mayaguez 270.....	78.72	16.2	9.86	41.92	17.4	5.66	120.64	15.52	3.83	.493
Mayaguez 275.....	66.84	15.0	7.71	49.93	16.1	6.14	116.77	13.85	3.71	.440
Mayaguez 317.....	66.32	15.5	7.95	44.96	16.5	5.71	111.28	13.66	3.53	.434
P. O. J. 2878.....	61.72	15.5	7.35	44.21	16.6	5.61	105.93	12.96	3.36	.411

¹ Planted Aug. 11, 1937; harvested Feb. 28, 1939.

² Harvested Mar. 27, 1940.

Because they produced more cane per acre, Mayaguez 275 and Mayaguez 317 yielded more sugar per acre than Mayaguez 270 in the first-ratoon crop, even though the sucrose content of the juice of

Mayaguez 270 was the highest in the test. As in the plant crop, all the Mayaguez varieties yielded more sugar per acre than either P. O. J. 2878 or F. C. 916.

Totaled for two crops, Mayaguez 270 produced the highest cane and sugar yields.

The total amounts of cane and of sugar produced by the varieties in the two crops are also presented in table 23. On this basis, Mayaguez 270 not only produced more cane per acre, but, because of the high sucrose content of its juice, also produced more sugar per acre than any of the other varieties. This variety averaged almost one-half of a ton of sugar per acre per month for the total 31½ months of combined plant and first-ratoon crops.

Seed pieces of new promising varieties were distributed.

The station has maintained small plats of a number of the varieties which have given promising yields in field experiments, in order to have available a supply of seed pieces for interested planters. During the year 60,775 seed pieces were distributed to 96 growers of the island requesting propagating material of such new varieties.

The sugarcane varieties Mayaguez 317 and Mayaguez 275 have been in great demand, a total of 21,626 seed pieces of these 2 varieties having been distributed.

Variety Mayaguez 338 showed susceptibility to *Fusarium* "pokkah boeng."

In the last few years the sugarcane variety Mayaguez 338, a progeny of the two varieties Mayaguez 28 and P. O. J. 2878, has been considerably extended in acreage; it not only yields good cane tonnage and juices, but because of its liberal stooling habit and ability to close in quickly between the rows, cultivation costs are low. It is also resistant to mosaic and the usual leaf spot diseases.

During the year, in a nursery field for the propagation of seed pieces, Mayaguez 338 was observed to be seriously affected with *Fusarium* pokkah boeng disease; the bases of the leaf blades of affected plants were characterized by being narrowed and having chlorotic patches. Such leaves were also distorted and had brownish tattered edges. Crop cane of the same variety in nearby fields showed scarcely any evidence of the disease. There may be significance in the correlation of recent and heavy applications of nitrogen fertilizers to the nursery field, whereas the crop cane had been fertilized but normally and not recently. It is possible that the avoidance of heavy fertilizer applications previous to the expectation of wet weather favorable for *Fusarium* development would be a sufficient measure to avoid pokkah boeng in this variety.

The sugarcane work has been in charge of Arthur G. Kevorkian, assistant plant pathologist and physiologist, during the year.

PLANT INTRODUCTIONS AND DISTRIBUTIONS

Brewers' yeast stimulated development of mangosteen seedlings.

The mangosteen (*Garcinia mangostana* L.), is reputed to be the most delicious of tropical fruits. Although the two bearing trees on the station grounds produce viable seed, until recently little headway

had been made in propagating this crop. Seeds germinated readily and the young seedlings grew vigorously until the stored food materials in the seeds were exhausted. When plants were grown in fertile potting soil, their root systems at this stage of development were poorly developed and most of the roots had a dead, corky outer covering and apparently inactive root tips. Frequently little or no growth was made after this stage, and most of the plants eventually died.

During the fiscal year 1939 it was found that when the substratum in which the mangosteen seedlings were grown was irrigated with a complete nutrient solution a much more vigorous growth resulted. Furthermore, an additional stimulation was obtained in the early



FIGURE 20.—Twenty-month-old mangosteen seedlings. *A*, Grown in dead sphagnum moss and given complete nutrients. *B*, grown in fertile seedbed without added nutrients. One-fourth natural size.

seedling stages when the irrigation solution contained a 1–1,000,000 water extract of brewers' yeast. In the early stage of the development of the seedlings a 75-percent increase in leaf area was obtained in the yeast-extract-fed plants over the non-yeast-fed plants.

A mangosteen orchard has been planted at the experiment station.

During the past fiscal year the yeast-fed plants continued to be larger than those that received no yeast, but the relative difference in size has become less as the non-yeast-fed plants passed the early critical growth period. However, the water extract of brewers' yeast was of practical importance in carrying the seedlings past the critical growth period several months after planting. Comparable seedlings of the same age which have been grown in fertile loam soil have developed only two or three pairs of small leaves in 20 months, and most of the plants have died. Plants that received the

nutrient solution and those that received the nutrient solution plus the yeast extract are from 12 to 18 inches tall and have begun to develop lateral branches. Comparative development of treated and untreated plants is shown in figure 20.

An article, giving in detail the results of this nutrition study with mangosteen seedlings, has been submitted and approved for publication in the *Journal of Agricultural Research*.

This improved technique in the propagation of mangosteens has made possible rapid strides in the increase of station plantings devoted to this crop. Thirty of the largest plants have been planted in an orchard in the best alluvial soil of the station. An additional 65 plants, which have been shifted to larger containers and are growing in the greenhouse under more carefully controlled conditions, will soon be large enough for planting in the orchard.

Mature mangosteen trees at station bore unusually heavy crop of fruit in 1940.

The two bearing mangosteen trees on the station grounds frequently bear two crops of fruit a year, a main crop which matures from June until October and a smaller crop which matures during the winter months. No fruit was borne on either of the two trees during the past winter; however, an unusually heavy summer crop followed, a total of more than 2,800 fruits maturing on the two trees. The largest previous yield of these two trees was 657 fruits during the summer of 1936. During previous seasons practically all flowers that opened developed into mature fruits. During the past season, of the total of 4,081 blossoms that opened on the two trees, only 2,818, or 69.1 percent, developed into mature fruits; 30.9 percent abscised soon after blossoming or later as immature fruits.

The unusually heavy crop resulted in fruit that was only approximately one-half the size produced in former years. However, the heavy yield and smallness of the fruits were in no way detrimental to the fine edible quality of the flesh and did not adversely affect seed formation within individual fruits.

During the past year bats were found to be damaging the mangosteen fruits. Of the 2,818 fruits harvested, 1,006, or 35.7 percent, were damaged by bats. The bats consumed the seeds as well as the succulent flesh and consequently reduced the amount of propagating material available. Mangosteen fruits that have been eaten by bats are shown in figure 21.

Mangosteen seeds differ from ordinary dicotyledonous seeds.

The mangosteen seed is so different in structure from ordinary dicotyledonous seeds that it has been termed a hypocotyl-tubercle. The mature seed consists of a central stelelike body surrounded by a fatty storage tissue. The tissue immediately surrounding the stelelike body was found to contain 63 percent of ether-extractable substances, whereas the value for the seed as a whole was 40 percent.

In a mature mangosteen seed no leaves, stem, or roots are present as in the usual seed; these develop after planting. First, a wartlike growth appears at a point on the seed surface. The location of this wartlike primordium is so extremely variable that it cannot be accurately predicted. The first root develops from this growth. Al-

most simultaneously, or shortly afterwards, another primordium develops at a point on the seed surface directly opposite the first, the shoot developing from this second growth point. When the shoot



FIGURE 21.—Fruits of *Garcinia mangostana* partially eaten by bats.

has developed to a length of approximately three-fourths of an inch a second root develops from the base of the shoot but not from the seed itself. The first root, which develops from the seed, usu-

ally becomes nonfunctional and withers away when the reserve food stored in the seed becomes exhausted. The small plant is thus equipped with a permanent root system, none of which originated directly from the seed itself. A typical germinated mangosteen seed is shown in figure 22.

Mangosteen seed germination was good.

Nearly all mangosteen seeds germinate without difficulty within 6 to 8 weeks after planting. The normal color of a young leaf is dull red which slowly turns to dark green in maturity. The leaf form changes between the first to about the fifth pair of leaves from cordate for the juvenile to large, acuminate for the typical adult form. The leaves are produced in close succession until the seed storage tissue is exhausted. The small plant is vigorous to this point.

In Mayaguez soil the plant loses its vigor as the seed food reserves are exhausted. At this stage of development the plant has few lateral

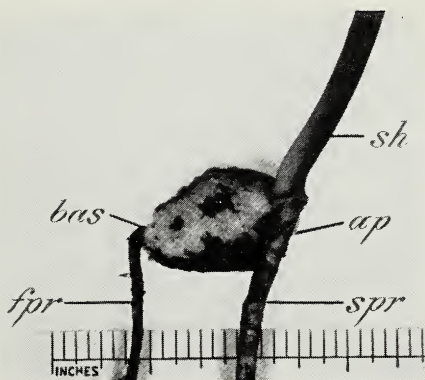


FIGURE 22.—Typical germinated seed of *Garcinia mangostana*; *bas*, Basal end; *ap*, apical end; *fpr*, first primary root; *spr*, second primary root from the base of *sh*, shoot.

roots, and most roots have a dead, corky outer covering and apparently inactive root tips. Plants fed with yeast and nutrient solution, however, have an extensive, active root system with little corky covering.

Mangosteen seed can form another shoot when original shoot dies.

When the shoot of a seedling dies as a result of transplanting or other causes before the food reserves in the seed are exhausted, a new shoot will often develop from the seed. In such cases several shoot primordia develop about the base of the dead original shoot and often more than one of these primordia develop into shoots. These new shoots appear to be equal in all respects to the original. An example of new shoots developing from the seed near the base of an original shoot that failed to continue growth is shown in figure 23.

Some mangosteen seeds are polyembryonic.

Frequently two, rarely three, and even more rarely four mangosteen plants have been observed to originate from a single seed. In cases of such polyembryony the embryos are tightly fitted together within a single fragile seed coat. Although it requires firm manipulation to separate the individual embryos prior to germination, each is a dis-

inct unit and all separate readily on germination. An example of mangosteen polyembryony is shown in figure 24.

Internal structure of mangosteen seed is similar to that of a tuber.

A study of the internal structure of a mature mangosteen seed showed close similarity to the form of a tuber in that the vascular system was interspersed with copious storage tissue; however, no "eyes" were present over the surface as in the case of a typical tuber. The tuber-like structure of the seed, together with the fact that multiple shoots



FIGURE 23.—Regerminated mangosteen seed. The growth failure of the central stein was followed by the development of two other shoots.

can originate from a whole monoembryonic seed, prompted a study of the behavior of fractioned mangosteen seeds.

Pieces from subdivided mangosteen seed produced normal plants.

Normal plants developed from embryo fractions obtained from seeds cut transversely into two and into three pieces, when the fractioned seeds were planted in dead sphagnum moss and given the same treatment as a whole seed.

Although the outline of the mangosteen seed is variable, the form most frequently encountered tends to be rounded at one end and pointed at the other. When germinated, the first primary root often develops from the pointed end and the shoot from the opposite end. When seeds of this form were cut transversely and planted, the pieces showed this same type of polarity. Shoots developed at the end of the seed

piece nearest that end of the intact seed which would normally produce a shoot, and roots developed on that portion of the fractioned embryo that was nearest the end of the seed which would normally produce a root. This was true whether the seed was cut into two or into three pieces. When a shoot developed on a fractioned embryo at a point where no shoot primordium existed before the seed was sectioned, the primordium giving rise to the shoot developed outside the periphery of the apical cut surface and never on the cut surface. Sometimes several shoots developed simultaneously from the same seed piece. New root primordia, on the other hand, originated on the cut surface itself from the periphery of the stelelike vascular system.

The ability of fractioned mangosteen seeds to produce normal plants is of important practical value under conditions where the supply of seed is limited and rapid expansion of plantings is desired. This type of propagation is being utilized to extend station plantings of this



FIGURE 24.—Triembryonic seed of *Garcinia mangostana*: A, Undivided seed showing snugly fitting embryos; B, the same seed with the three embryos separated.

valuable new fruit crop for the island. A typical germinated trisected mangosteen seed is shown in figure 25.

Many new plant species were received during the year.

Through exchanges, gifts, and the continued close cooperation of the Division of Plant Exploration and Introduction of the Bureau of Plant Industry, a total of 571 new plant species was received during the year. Of the new species, 10 were classified as drug plants, 10 sugarcane, fiber, and forage crops, 31 fruit plants, 13 insecticidal plants, 13 perfume and spice plants, 8 timber plants, 157 ornamentals, 52 palms, and 277 orchids. Species of special interest are varieties of the fruit tree, sapote, from Colombia and Ecuador, citronella grass from Java, tonkabean from Colombia, a true white-flowered form of *Bougainvillea glabra* Choisy from Brazil, the walkingstick palm from the Solomon Islands, and the drug-yielding *Strychnos toxifera* Schomb. ex Benth., from Panama.

Many of the new plants received during the year, particularly new orchid species, were obtained by Director Lee, while assigned by the



FIGURE 25.—Germinated trisected seed of *Garcinia mangostana*: A, Apical-end section; B, midsection; C, basal-end section: *apcs*, Apical cut surface; *appr*, apical primordia; *bcs*, basal cut surface; *fpr*, first primary root; *sh*, shoot; *spr*, second primary root; *ss*, secondary shoot; *st*, stelelike body.

United States Government to work as Agricultural Adviser in Haiti, Ecuador, and Colombia.

Progress was made in orchid-culture experimentation.

Interest in orchid culture in Puerto Rico has recently become intensified by the assembling of a large collection of species by the station for experimental purposes. Experimental work with orchid culture in



FIGURE 26.—*Calamus* sp. in the newly established palmetum.

Puerto Rico is conducted with the object of developing a cut-flower industry to supply the eastern United States market, shipment being made by air. During the last 2 years a collection consisting of 150 named species of orchids has been assembled from Brazil, Trinidad, Venezuela, Costa Rica, Guatemala, Honduras, Panama, Colombia, and

Ecuador, and also from the United States. Many other unidentified species were received. As soon as the doubtful species produced flowers, specimens, descriptions, photographs of the plant and other material were sent to Charles Schweinfurth, of Harvard University, who has identified many of the newly received species.

Rattan cane-yielding palmetum shows adaptability of these commercially valuable palms.

The rattans are Asiatic or African climbing palms and practically all commercial rattan canes have been imported from the oriental Tropics. Rarely is a palm of this tribe found in American botanical gardens. During recent years a rattan palmetum has been established



FIGURE 27.—Young fruiting tree of *Aleurites trisperma* growing as an avenue tree in Cabo Rojo.

at this station. The rapid growth made by these palms, illustrated in figure 26, shows that they are well adapted to forest culture in this environment. Plants have been grown from seed of species from the Philippines, India, Malaya, and Sumatra, and plantings are now well established. To date, attempts to germinate seeds of the African spineless rattan palms of the genus *Ancistrophyllum* have failed.

A satisfactory method of packing palm seed for distant transportation was developed.

Experience at the station indicated that the failure of these palm seeds to germinate may have been the result of improper packing of the seeds at their source.

Seeds of most palm species retain viability only if the moisture about them is near a definite level. The most frequent packing error is to use too much moisture. Under this condition the seeds germinate and diseases often kill the new plants before they reach their destination. A less frequent error in packing is to use packing medium that has too little moisture,

During the past few years an effective method of packing palm seed for distant shipment was developed. Excellent results were obtained when the seeds were packed in a medium of granulated peat or the dusty, granular, corky material from coir with a moisture content of approximately 30 percent. The mixture of seeds and packing material was hermetically sealed in ordinary tin cans such as those used in the preservation of fruits and vegetables. Any airtight container which would maintain the moisture content of the packing material until the seeds reached their destination would doubtless be satisfac-



FIGURE 28.—A rubber tree, *Hevea brasiliensis*, at Mayaguez, approximately 30 years old.

tory. If the proper moisture content has been obtained for the packing material and the tin container is properly sealed, the palm seeds cannot lose sufficient moisture to lower their viability and they cannot absorb sufficient moisture to germinate during transit or storage. This type of packing has proved most satisfactory for shipments of many other kinds of seeds also.

Philippine paint-oil tree has shown further adaptability to Puerto Rican conditions.

Aleurites trisperma, the Philippine paint-oil tree, has continued to show promise as a new crop. It now appears certain that this

species can be depended upon for successful growth in Puerto Rico and thus give a domestic source of the tunglike oil so essential in the continental paint industry. The tree grows well at Mayaguez, where the annual rainfall is 89 inches, and also thrives as an avenue tree in the town of Cabo Rojo, where the average annual rainfall is only 65 inches. One of the trees in Cabo Rojo is shown in figure 27.

Seeds from individual trees growing on the station grounds were harvested for examination by the Bureau of Agricultural Chemistry and Engineering to determine if there was much variation in oil quality between trees. Should considerable variation in oil quality be found, studies of methods of propagating the species asexually could be undertaken in an effort to propagate as clones the most desirable specimens.

Rubber grew well at Mayaguez.

Plants of *Hevea brasiliensis* (H. B. K.) Muell. Arg. were among those first sent to this station by the Division of Plant Exploration and Introduction of the Bureau of Plant Industry, soon after its establishment in 1902. Other lots of propagating material have been obtained from time to time. At present about 30 mature trees are growing in different locations, including various soil and moisture conditions provided on the station grounds. Because of the limited land areas and the hurricane liability, it has been felt that rubber could not be developed as a profitable crop in Puerto Rico, and hence no intensive research with rubber production has been conducted here. The plants grow rapidly and form large trees, as is shown in figure 28. No systematic tappings have been made to determine quality and yields, but occasional tappings have shown that the latex yield is profuse.

Propagating material of more than 81,000 plants was distributed for trial.

During the past year a total of more than 81,000 cuttings and plants were distributed to interested persons in all sections of the island for trial. Although the propagating material distributed consisted mostly of ornamental species, it included also more than 14,000 seedlings of improved mango varieties and more than 1,000 seedlings of avocado. The propagating material of the different types of plants distributed during the year is shown by months in table 24.

TABLE 24.—Economic and ornamental plants distributed during the fiscal year 1940

Kind	July	Aug.	Sept.	Oct.	Nov.	Dec.	Jan.	Feb.	Mar.	Apr.	May	June	Total
	No.	No.	No.	No.	No.	No.	No.	No.	No.	No.	No.	No.	No.
Avocado.....		276	350	12	249	4	77	11	29	24		11	1,043
Bamboo.....	38	62	10			129							239
Bougainvillea.....	156	197	145	70	128	187	150	251	462	273	268	417	2,704
Cactus.....	106	129	76	38	27		101	28	44	32	27	68	676
Citrus.....	50					4		6	11	19			90
Hibiscus.....	1,140	1,624	4,409	1,268	3,172	6,710	1,540	3,002	3,291	4,041	3,480	3,157	36,825
Mango.....	184		1,837	7,636	2,662	109	287	12	73	53	11	1,537	14,401
Miscellaneous economic plants.....	44	299	298	192	5,052	523	22	24	6	32	120	821	7,433
Miscellaneous fruit plants.....	136	45	81	100	6	59	61	72	6	78	26	36	706
Miscellaneous ornamental plants.....	111	237	147	124		162	111	190		148	50	54	1,334
Ornamental shrubs.....	530	371	304	274	1,060	1,272	661	129	170	371	828	219	6,169
Ornamental trees.....	412	127	385	113	1,232	368	233	127	112	196	498	244	4,047
Ornamental vines.....	508	250	97	74	210	119	130	104	100	85	365	474	2,516
Palms.....	130	240	152	112	1,033	239	136	61	159	20	195	57	2,546
Waterlilies.....	42	9	12	9	39	2		14	18	30	34	14	223
Wax flowers.....	9	8	31	13		12	16	53	105	47		37	334
Total.....	3,596	3,874	8,328	10,035	14,870	9,899	3,525	4,087	4,566	5,458	5,902	7,146	81,286

C. L. Horn, associate horticulturist, was in charge of plant introductions and distributions during the year.

BIOLOGICAL-CONTROL ACTIVITIES

Five new species of beneficial insects were introduced during the year.

During the year 5 new species of beneficial insects were introduced into Puerto Rico for the station, bringing the total to 75 species since the inception of the biological-control program. Of this number 17 species are known to be successfully colonized and in many instances are aiding considerably in the control of their particular host pests.

Cooperation with other countries of the Western Hemisphere in the exchange of insect parasites and predators was continued, and material was also sent to and received from the continental United States. The Division of Foreign Parasite Introduction of the Bureau of Entomology and Plant Quarantine took an active part in directing the work in progress and in arranging for the importation of black scale parasites from the Citrus Experiment Station of the University of California. A shipment of coccinellid beetles was also received from Brazil through the cooperation of the Instituto Agrônômico do Estado de São Paulo, Campinas, Brazil, and sugarcane borer parasites were received from the Department of Agriculture, Barbados.

The results obtained thus far from previous introductions and the colonization and rearing of the newly introduced species are briefly reviewed as follows:

Diatraea borers for rearing parasites were provided through the cooperation of local sugarcane growers.

The rearing of *Metagonistylum minense* Tns. and *Theresia clari-palpis* (V. D. W.), fly parasites that attack the sugarcane borer (*Diatraea saccharalis* (F.)), was carried on during the year through the cooperation of the following sugarcane growers: Asociación Azucarera Cooperativa Lafayette, the Fajardo Sugar Co., Luce & Co., Russell & Co., and Sucesión J. Serrallés. *Diatraea* borers were collected in the field by these growers and sent to Mayaguez where they were used in rearing sugarcane borer parasites.

Two strains of *Metagonistylum minense*, the Amazon fly, have been introduced.

The occurrence of two physiological strains of *Metagonistylum minense* was discussed in the 1939 annual report. One of these, native to the Amazon River region, is a strain adapted to wet regions, while the other, obtained at São Paulo, Brazil, is adapted to a comparatively dry climate. The latter, or São Paulo, strain has been considered by Harland (?) to be melanic. Observations made on this strain showed that in a small percentage of the material reared the abdomen had a distinct reddish cast as opposed to the distinctly characteristic black coloration expected. This variation in color was looked for in material of the São Paulo strain received from Barbados during July, but all of the mated females received were found to be melanic. However, progeny reared from these females produced a few specimens having a reddish cast on the abdomen.

When flies of the São Paulo strain having the reddish cast were mated, their progeny proved to be predominantly black, but a small

percentage always showed a reddish coloration of the abdomen. It would appear from these rearings that both a melanic and a reddish form occur in the State of São Paulo, Brazil, although the melanic appears to be the dominant type.

The two strains readily cross and variations in the color markings result. Tucker (15) has discussed numerous crosses made by him between the two strains, and he notes a difference in fecundity in addition to color variations. Our observations would indicate that this difference is due to the length of gestation period necessary for the maximum development of larvae. Under our conditions, the São Paulo strain requires 24 to 48 hours more time for comparative development of larvae within the uterus than the Amazon strain.

Through the cooperation of R. W. E. Tucker, entomologist, Department of Science and Agriculture, Barbados, the station obtained for rearing purposes 22 mated females of the São Paulo strain collected by Mr. Tucker in São Paulo and also 23 mated females from St. Lucia obtained from British Guiana and later established and acclimatized in St. Lucia.

The inoculations and rearing data on the various strains of *M. minense* carried on during the year are shown in table 25.

TABLE 25.—*The inoculation of Diatraea saccharalis borers for the rearing of the various strains of Metagonistylum minense, giving numbers of each inoculated and adult flies reared*

Strain of <i>Metagonistylum minense</i>	<i>Diatraea saccharalis</i> larvae inoculated	Flies reared	
	Number	Number	Percent
Amazon.....	3,299	1,278	38.7
Amazon (St. Lucia).....	5,314	2,045	38.5
São Paulo.....	36,238	12,895	35.6
Total.....	44,851	16,218	36.2

Liberations of *Metagonistylum minense* were made in various sections of the island.

The physiological strains of *Metagonistylum minense* were liberated in various sugarcane-growing areas of the island as shown in table 26.

TABLE 26.—*The liberation of adult flies of the various physiological strains of Metagonistylum minense, giving locations and number of flies liberated between July 1, 1939, and June 30, 1940*

Locations	Amazon	Amazon (St. Lucia)	São Paulo	Total
	Number	Number	Number	Number
Arroyo.....	193			193
Cabo Rojo.....	450	3,124		3,574
Fajardo.....		546		546
Guayama.....		4,087		4,087
Ponce.....		1,835		1,835
Santa Isabel.....	118		1,626	1,744
Toa Baia.....		152		152
Yabucoa.....	107			107
Total.....	868	9,744	1,626	12,238

Both strains of *Metagonistylum minense* were recovered.

Throughout the year collections of *Diatraea saccharalis* borers were made in the vicinity of a number of the points where parasites of this sugarcane pest were liberated. The only newly introduced parasite found was *Metagonistylum minense*. Specimens of the native fly parasite, *Lixophaga diatraea* Tns., and also specimens of a previously introduced wasp, *Bassus stigmaterus* (Cress.), were found parasitizing the *Diatraea* borers collected.

Recoveries of *Metagonistylum minense* were made at Cabo Rojo, Fajardo, Guayama, and Santa Isabel, but in all cases the percentage of parasitization was low. The highest found was at Cabo Rojo where 6.9 percent of the borers collected were found to be parasitized.

From the recoveries made it appears that the Amazon strain of *Metagonistylum minense* is established on the south coast of Puerto Rico and that considerable spread has taken place during the past year. However, the percentage of parasitization remains low, and as yet this beneficial insect is of little importance in the control of the sugarcane borer.

The São Paulo strain of *Metagonistylum minense* recently introduced from Brazil and believed to be more adapted to the dry environmental conditions of the south coast is apparently becoming established and, after 1 or 2 years, may prove to be of more value than the Amazon strain.

Shipments of *Metagonistylum minense* were made to Louisiana and Barbados.

At the request of C. P. Clausen, in charge of Foreign Parasite Introduction of the Bureau of Entomology and Plant Quarantine, a shipment of 32 mated females of the São Paulo strain of *Metagonistylum minense* was sent to J. W. Ingram of the same Bureau at Houma, La. A shipment of 24 mated females of the same strain was sent to R. W. E. Tucker, entomologist, Department of Science and Agriculture, Barbados, upon his request, for study and comparison with his own material from the same habitat. Mr. Tucker reported that 18 flies were alive upon arrival.

A shipment of 22 mated females of the Amazon strain of *Metagonistylum minense* was also made to Barbados. This shipment was received in good condition, with 3 flies dead on arrival.

Liberations of *Theresia claripalpis*, sugarcane borer parasite, were made at Guayama.

The rearing of *Theresia claripalpis*, a parasite of *Diatraea saccharalis*, was continued for a considerable part of the year. A total of 4,637 sugarcane borers was inoculated, from which 1,249, or 26.9 percent, flies emerged.

Liberations of 496 adult flies of *T. claripalpis* were made at Guayama during the year. Two factors have limited the mass rearing and liberation of this parasite, the difficulty of mating and the high mortality during the gestation period which takes from 13 to 15 days depending upon the season of year. A highly satisfactory mating technique was finally perfected which resulted in practically every female being mated with a minimum of time expended.

The males and females were separated at the time of emergence or shortly thereafter. The males were held for at least 48 hours be-

fore attempting to use them for mating purposes. At the time of mating the cages containing the two sexes were placed in a bright light and allowed to remain there until the males became very active and were observed to be darting rapidly about. By placing an individual active male with a female in a narrow 2-inch glass vial, copulation usually resulted almost at once. Practically every female was readily and easily mated in this way.

A shipment of *Coelophora inaequalis*, predator of yellow sugarcane aphid, was sent to Louisiana.

The yellow sugarcane aphid (*Sipha flava* Forbes) is a serious pest of sugarcane throughout the West Indies, northern South America, and the southern continental United States. As reported in the 1938 annual report, a coccinellid beetle, *Coelophora inaequalis* (F.), was introduced from Hawaii for liberation against this cane pest.

During the year a small breeding stock of this predaceous beetle has been maintained, and liberations were made as follows: Fajardo 109, Guayama 111, Lajas 121, Mayaguez 108, San German 335, and Villalba 285.

During July a shipment of 100 adult beetles of *Coelophora inaequalis* was made by air express to J. W. Ingram of the Bureau of Entomology and Plant Quarantine at Houma, La., for liberation against *Sipha flava* in the continental United States.

Palm mealybug parasite effectively reduced population of *Pseudococcus nipae*.

The palm mealybug (*Pseudococcus nipae* (Mask.)) is a pest of avocados, guavas, palms, and other ornamentals. As reported in the annual report for 1939, there was introduced into Puerto Rico from Hawaii a parasite, *Pseudophycus utilis* Timb., which shortly after its liberation showed excellent promise of establishment. During the past year the extent of control exerted by this parasite has increased to the point where it is extremely difficult to find host material in those areas where liberations were originally made.

During the year collections of parasitized material have been distributed in new areas as follows: Añasco, Cabo Rojo, Fajardo, Las Mesas in Mayaguez, Naguabo, and San German.

From the results obtained thus far there is every indication that this mealybug will be brought under complete control by this introduced parasite.

Reduction in pineapple mealybug infestation was notable in Lajas district.

The establishment of *Hambletonia pseudococcina* Comp., a parasite of the pineapple mealybug (*Pseudococcus brevipes* (Ckll.)), in the district about Lajas has resulted in a healthier stand of pineapple plants than has been known for many years in that district. The yellowish, flaccid plants common a few years ago are now rarely seen. The mealybug population, which was exceptionally high, has been notably reduced. It is of importance to note that in other districts, such as Arecibo and Corozal, where *H. pseudococcina* is also established, there has not been the same apparent reduction in the mealybug population. This has probably been influenced by such factors as environment, type of cultivation, and the original concentration of host population.

A small breeding stock of *Anagyrus coccidivorus* Dozier, another introduced pineapple mealybug parasite, was maintained during the year, and liberations of the species were made at Bayamon, 93 adults, and Lajas, 502 adults. In addition, from field recoveries incidental to the above rearing work, liberations of *H. pseudococcina* adults were made as follows: Bayamon 47, and Mayaguez 85.

A few recoveries of *A. coccidivorus* were made in the vicinity of the liberation point at Lajas, but this species cannot be considered as well established at this time.

Egius platycephalus, bamboo scale predator, is doing effective work.

One of the most common predators to be found feeding on the bamboo scales (*Asterolecanium bambusae* Bdv. and *A. miliaris* Bdv.) is a ladybeetle, *Egius platycephalus* Muls., introduced from Cuba in 1938. Large numbers of this coccinellid were observed throughout the island. In many instances it was possible to date the establishment of *Egius* on a particular bamboo clump by the clean appearance of the culms, free from bamboo scale, which had grown out in the past 1, 2, or 3 years, as the case might be. In some areas this reduction in scale has been brought about also by the help of other introduced scale predators which are subsequently discussed.

Numerous scale predators are now well established in various sections of the island.

A bamboo scale predator, the coccinellid *Cladis nitidula* F., introduced from Martinique in 1938 and previously known as *Curinus* sp., is now well established in various sections of the island. This species is largely found attacking *Asterolecanium bambusae* which is common on the culms of bamboo.

Curinus sp. introduced from Trinidad in 1938 is also well established in various localities where it is found feeding commonly on *A. miliaris*, common on the leaves of bamboo.

Pentilia castanea Muls., an introduction from Trinidad in 1938, is well established in many localities. This ladybeetle not only feeds on the bamboo scales but is also commonly found on the white scale (*Aulacaspis pentagona* Targ.) of papaya and also on various citrus scales.

Chilocorus cacti continues as effective control of white papaya scale.

The coccinellid predator *Chilocorus cacti* L., introduced from Texas and Louisiana in 1937 and from Cuba in 1938, has proved to be particularly effective in the control of white peach scale (*Aulacaspis pentagona*) on papaya. Large colonies have been observed at Arecibo, Bayamon, Isabela, and Mayaguez, in many instances several miles from the nearest liberation point.

Heavy infestations of scale are apparently preferred by this species and they tend to disperse or die out rapidly as the scale population diminishes. Their ability to seek out heavy scale infestations and to build up rapidly thereafter, however, makes them of considerable value and importance in the control of this papaya scale pest.

New bamboo scale predator was received from Brazil.

A shipment of predatory ladybeetles, *Exochomus jourdanii* Muls., was received from Felisberto Camargo of the Instituto Agrônômico do Estado de São Paulo in June. These beetles were collected at

Santos, Brazil, where they were found feeding on the bamboo scales (*Asterolecanium* spp.).

The shipment of beetles was made by air express in tin cans fitted with a roosting surface of mesh wire. Between the meshes were placed a number of cotton dental wicks soaked in water to provide a high humidity. A previous attempt to introduce this species was made in June 1939, but because of the type of shipping container used all of the material arrived dead. In the last shipment received 449 beetles of 711 shipped were alive on arrival.

A few of the beetles received were retained for rearing and were placed in cloth spindle cages set around bamboo culms infested with the scale *Asterolecanium bambusae*. A total of 45 beetles was placed in three cages; at the end of June some reproduction had taken place and all stages of the beetles were to be found. The cloth bags were then left open at the top to permit the gradual escape of the new generation of beetles.

Scale predators were redistributed to new localities.

Liberations of *Egus platycephalus* were made at Arecibo in September and at Las Ochenta in Mayaguez. *Chilocorus cacti* was redistributed at Las Mesas in Mayaguez in March. *Pentilia castanea* was liberated at Isabela in September. The newly introduced predator of bamboo scales, *Exochomus jourdani*, was liberated at Mayaguez in June.

Shipment of *Egus platycephalus* was made to Haiti.

On September 23 a collection of 300 adults of *Egus platycephalus* was made on bamboo at Mayaguez. These bamboo scale predators were taken to Haiti as personal baggage by Georges Heraux, Agronome en Chef of the Service Technique of the Government of Haiti. The beetles were reported to have arrived in excellent condition and were liberated about Port-au-Prince, Haiti.

Black scale parasites were introduced from California.

Shipments of four species of parasites which attack the black scale (*Saissetia oleae* (Bern.)) were received in June from H. S. Smith of the Citrus Experiment Station, Riverside, Calif.

Rearing work with *Diversinervus* sp., *Mesopletis* sp., *Metaphycus helvohus* Comp., and *Metaphycus stanleyi* Comp., all African parasites of *S. oleae*, was started.

A liberation of 50 adults of *Mesopletis* sp. was made on the station grounds at Mayaguez in June.

Shipment of negro scale was made to California.

During May a shipment of locally collected negro scale (*Saissetia nigra* Niet.) was sent to the Citrus Experiment Station at Riverside, Calif. S. E. Flanders of that station, who received the material, reported that the following parasites and predators were reared: *Scutellista cyanea* Motsch., *Eupelmus coccidivorus* Gahan, *Lecanobius cockerelli* Ash., *Coccophagus scutellaris* Dahm., *Aneristus ceroplastae* How., *Marietta* sp., and *Scymnus flavifrons* Melsh.

Shipment of *Spalangia* sp. was sent to Colombia to aid in the control of the horn fly of cattle.

During April a request was received from the Federal Department of Agriculture of the Republic of Colombia for parasites to aid in the

control of the horn fly (*Haematobia irritans* L.) of cattle. Rearing material for this shipment was obtained from field collections of *H. irritans* puparia made in those locations where the introduced parasite *Spalangia philippinensis* (Full.) had previously been liberated.

The adults reared were used for further laboratory breeding, and on May 22 a shipment of 500 adults of *Spalangia philippinensis* was sent to Dr. Luis María Murillo, Chief of the Entomology Service of Colombia. The material was sent by air express, and Dr. Murillo reported that 497 parasites arrived alive.

Shipment of giant toad *Bufo marinus* L. was sent to Cuba.

At the request of L. C. Scaramuzza, in cooperation with C. P. Clausen of the Bureau of Entomology and Plant Quarantine, a shipment of 44 adult giant toads, *Bufo marinus*, was made by steamer to Cuba in May. The toads were packed in a wooden box on the bottom of which was placed a thick layer of coconut fiber soaked in water. Mr. Scaramuzza reported that the toads arrived in good condition and that 43 were alive on arrival.

Kenneth A. Bartlett, entomologist, was in charge of biological control activities during the past year.

ENTOMOLOGICAL INVESTIGATIONS

BIOLOGICAL STUDIES OF THE BAMBOO POWDER-POST BEETLE

Beetles tunneled most into soft, pithy wood on inside of bamboo culms.

While carrying out observations on the feeding habits of the powder-post beetle (*Dinoderus minutus* F.) in bamboo, it was noted that after the adults had entered the wood they usually tunneled into the softest parts of the culm. Ordinarily the softest part of solid culms is the center and of hollow culms the inner part of the wall. The fibrovascular bundles here are larger, contain larger vessels, and are spaced farther apart than those located toward the rind. The soft, pithy tissue that fills the intervening spaces between bundles is thus more abundant in the center of solid culms or in the inner part of the wall of hollow culms than toward the outside; usually this tissue is rich in starch. It has been observed that, other things being equal, the more of this spongy tissue in a piece of bamboo the heavier the attack by the powder-post beetle; a typical case is shown in figure 29.

Eggs were found in tubular vessels in inside wall of the culms.

While the beetles often entered longitudinally with the grain of the wood, as from a cut end, as shown in figure 30, *a*, within a short distance from the entrance they were observed to run their tunnels transversely, or at right angles, to the grain, as in figure 30, *b*. In pieces of bamboo that had been exposed for some time, these tunnels were often found extending all the way around the inside of the culm or just beneath the inner surface of the wall, figure 30, *c*.

In splitting open pieces of 1-year-old culms of the common hollow bamboo (*Bambusa vulgaris*) that had been exposed to infestation for about 6 weeks, many small larvae were found burrowing in the pithy tissue between the fibrovascular bundles close to transverse tunnels inhabited by adults, figure 30, *d*. The larvae occurred in greatest abun-

dance within about one-half inch of the outside entrance and from there gradually decreased in number and size. Further splitting longitudinally across the tunnels revealed that these larvae came from eggs deposited in the vessels of the fibrovascular bundles. One of the eggs found is shown in place in figure 31. Evidently the transverse tunnels were made for the purpose of oviposition, as eggs or newly

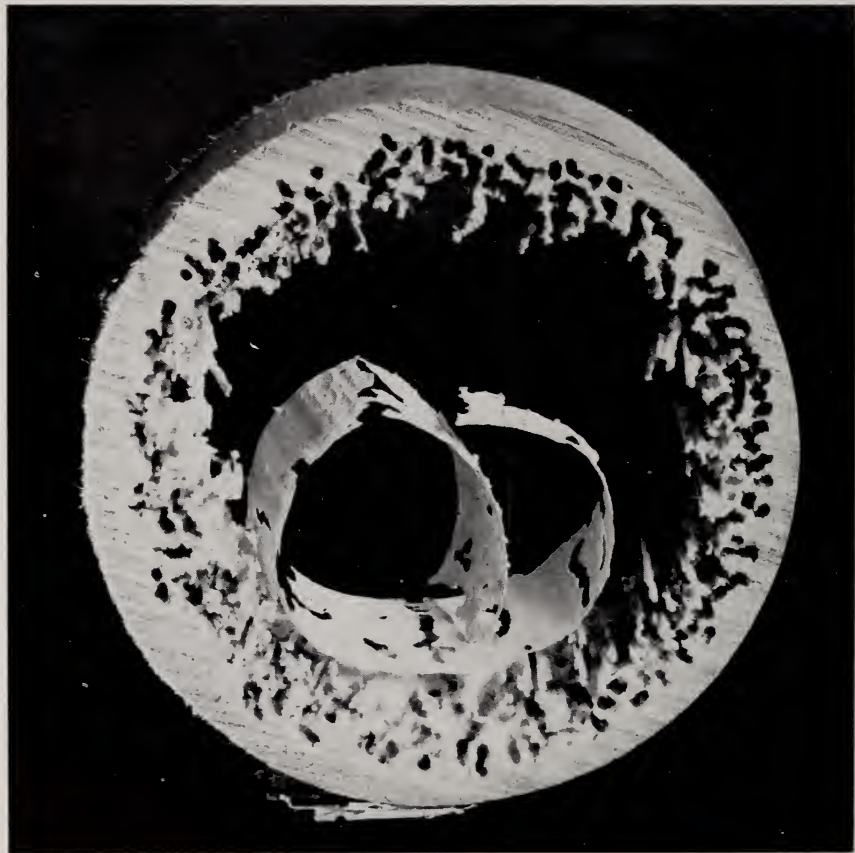


FIGURE 29.—Cross section of a basal internode of a 1-year-old culm of the common bamboo (*Bambusa vulgaris*) showing soft, pithy wood in the inner part of the wall eaten away by larvae and adults of the bamboo powder-post beetle (*Dinoderus minutus*). Before infestation, this part of the wall reacted strongly to the iodine test for starch. The parts remaining afterward consisted mostly of tough fibers and wood that reacted feebly to this test. \times about 1.25.

hatched larvae were found in sections from the entire length of such tunnels.

Eggs were inserted in ends of vessels severed during extension of oviposition tunnel.

While not actually observed, it was apparent that as the female continued to bore into the soft wood she inserted eggs into the ends of the severed vessels leading from the oviposition tunnel. Since no eggs could be found in other locations, such as in the exposed edges of a

cross section or similar cut, it is probable that this is the usual, if not the only, method of oviposition in bamboo. No more than one egg was ever found in the same vessel, unless the vessel were cut by more than one tunnel. The larger or more broadly rounded extremity of the egg was always inserted first, as is also shown in figure 31. The other extremity of the egg was usually about three-fourths of a millimeter from the tunnel end of the vessel, which was always closed with woody excrement, as shown in figure 32. Whether this closing of the vessel was part of the act of oviposition or a result of ordinary movements of the adult back and forth in the tunnel could not be definitely ascertained. However, it is probable that the former was the

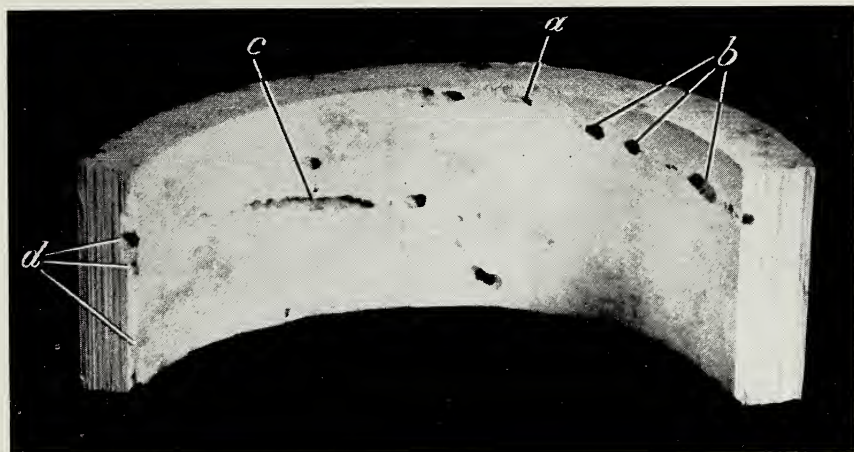


FIGURE 30.—Cross section of a 1-year-old culm of the common bamboo split to show oviposition tunnels made by the bamboo powder-post beetle. Entrance of one adult is shown at a point on the upper edge a little to the right of the middle (*a*). Note oviposition tunnel extending transversely to the grain to the right from this point around the inside of the culm wall (*b*). Another such tunnel is shown extending to the left (*c*), and its cross section is seen in the split edge at the extreme left. Above and below this tunnel, some of the soft, pithy wood in the inner part of the wall can be seen eaten away by larvae from eggs deposited in the vessels severed by the tunnel (*d*). \times about 1.5.

case, since there was more excrement and it seemed to be packed more tightly in the ends of vessels containing eggs than in others.

Beetles oviposited freely in loose excrement from sweet corn kernels.

Because of their extremely fragile nature, eggs could not be extracted in sufficient number from bamboo wood for any but casual observations. To secure data on oviposition and a supply of eggs for studies of the larval and pupal stages, other methods were tried.

By frequent examination of beetles taken from oviposition tunnels and caged with some of the powdery excrement from these tunnels, it was found that under these artificial conditions eggs were sometimes deposited loosely in this excrement. However, the beetles soon died, and the small number of eggs deposited seriously limited the practicability of this method when used in rearing work.

Since *Dinoderus minutus* is known to infest sweet corn (19, p. 242), dry kernels of the U. S. D. A.-34 variety, developed at the station, were tried as an oviposition medium. When caged in vials with several kernels of this corn partly split open, the adults began to feed almost immediately. The beetles seemed to live normally in this medium and soon began to oviposit freely in the excrement resulting from



FIGURE 31.—Egg of *Dinoderus minutus* in tubular vessel in wood of the common bamboo (*Bambusa vulgaris*). In splitting the wood, the usual plug of beetle excrement in the end of the vessel leading from the oviposition tunnel, at top of photograph, was removed, and part of the vessel lining was left over the egg. Note large end of egg inserted first. \times about 55.

boring and feeding. Each day the kernels of corn and the vials were cleaned, and the excrement was searched under the microscope for eggs. In this way it was possible to secure intact any desired number of eggs of known date of deposition for incubation and other biological studies. From eggs thus obtained the following data on oviposition and incubation were secured up to the end of June.

Between May 24 and June 16, 7 newly emerged females confined in a loosely stoppered vial with 7 males of approximately the same age and with split kernels of sweet corn for food, deposited 93 eggs. Dur-



FIGURE 32.—Piece of common bamboo split longitudinally to show egg of *Dinoderus minutus* in vessel in wood. Part of cross section of oviposition tunnel is shown at top. Note end of vessel plugged with excrement from floor of tunnel. \times about 55.

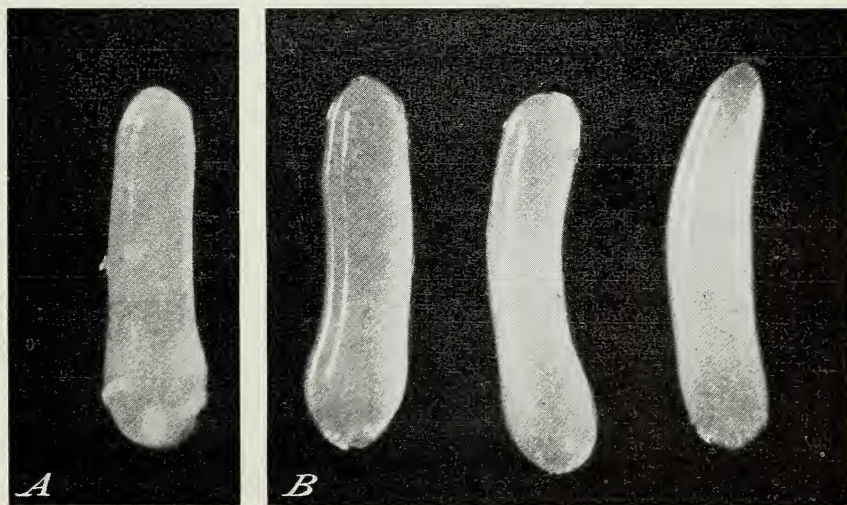


FIGURE 33.—Eggs of *Dinoderus minutus* of typical shapes and sizes: A, Egg taken from vessel in bamboo wood; \times about 65. B, three eggs found in excrement from sweet corn; \times about 70.

ing the remainder of June, 6 of these females caged singly on the same medium deposited 224 eggs, or an average from May 24 to June 30 of approximately 50 eggs per female. The largest number of eggs collected from a single female in any 1 day during this period was 14.

Eggs were elongate oval, slightly curved, and opalescent white.

The eggs found in bamboo did not vary greatly in shape or size from those deposited loosely in excrement from corn. A series of eggs deposited under both conditions is shown greatly enlarged in figure 33. Egg A was taken from bamboo wood and photographed 3 days before hatching. The shape and size of this egg and of those shown in figures 31 and 32 were typical of eggs found in this location. The other three eggs, B, from 1 to 3 days old at time of photographing, were typical of those found deposited loosely. As shown in figures 31 to 33, the eggs were elongate oval and somewhat curved to nearly straight, depending on whether they were inserted in tubular vessels in bamboo wood or deposited loosely in excrement. In both cases, one end was more sharply rounded than the other and sometimes tapered a little for about one-fourth the length of the egg.

The color of newly deposited eggs was opalescent white, changing gradually with age to a waxy white. The surface at first was shiny and without any apparent markings, but later became dull and sometimes slightly granular to wrinkled a day or two before hatching.

Newly deposited eggs averaged 0.81 millimeter in length by about 0.17 millimeter in diameter.

A number of eggs from both locations were measured under the microscope. Those found in bamboo wood were at about the middle of their incubation period, but those found in excrement had been deposited the day previously. These measurements are summarized in table 27.

TABLE 27.—Measurements of eggs of *Dinoderus minutus* deposited in bamboo wood and loosely in excrement from corn

8 EGGS DEPOSITED IN BAMBOO WOOD

	Length	Width		
		Broad end ¹	Middle	Narrow end ¹
Maximum.....	Mm. 0.89	Mm. 0.27		Mm. 0.18
Minimum.....	.75	.18		.17
Average.....	.81	.22		.18

32 EGGS DEPOSITED LOOSELY

Maximum.....	0.94	0.22	0.19	0.18
Minimum.....	.63	.15	.14	.11
Average.....	.81	.19	.17	.15

¹ Measured at about 0.10 mm. from end.

It will be noted in table 27 that the 8 eggs deposited naturally in bamboo wood averaged 0.81 millimeter in length, the same as the 32 eggs deposited loosely. However, the broad and narrow ends

of the former, which averaged 0.22 millimeter and 0.18 millimeter in diameter, respectively, were each 0.03 millimeter greater than the average widths of the latter.

Size of eggs diminished as incubation progressed.

Eggs kept in tightly stoppered vials were noted to decrease somewhat in size as incubation progressed. One egg found in excrement on May 8 measured 0.91 millimeter in length by 0.16 millimeter in middle diameter; 5 days later, just prior to hatching, this egg measured 0.85 by 0.15 millimeter, a shrinkage in both dimensions of well over 6 percent. Four other eggs, newly deposited or in early stages of incubation, averaged 0.81 millimeter in length by 0.18 millimeter in median diameter; when measured the day following, these

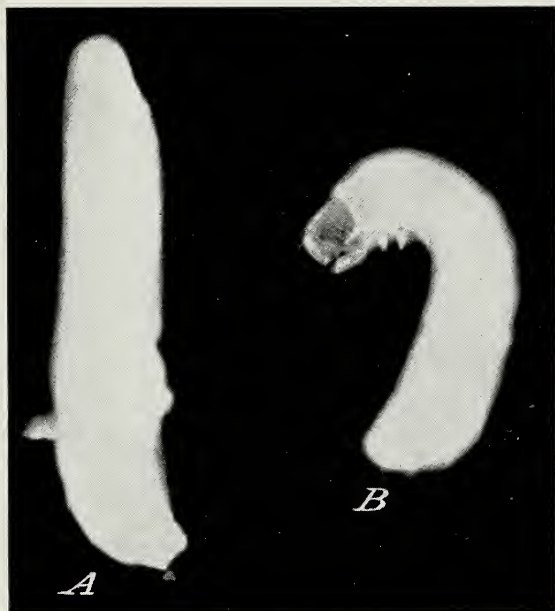


FIGURE 34.—Hatching and newly hatched larvae of *Dinoderus minutus*. A, Larva in the act of hatching. Note tip of abdomen protruding through break in large end of egg shell. B, A newly hatched larva. \times about 70.

eggs were found to have shrunk approximately 3 percent in length and 2 percent in diameter. All these eggs hatched into normal larvae.

Hatching occurred about 5 days after deposition.

Between the middle of May and the last of June incubation records were secured for 130 eggs. These eggs were caged singly without added moisture in ordinary 2-dram homeo vials fitted with cork stoppers and held at room temperatures, which averaged approximately 79° F. Under these conditions the incubation periods varied from 3 to 7 days and averaged 5.4 days for the 130 eggs observed.

In hatching it was noted that the larva first broke through the larger end of the egg shell with the tip of its abdomen. Sometimes this part of the shell was cut almost completely around so that it

remained fastened at one point like a lid. The larva then gradually backed out of the shell or sometimes freed itself by breaking through at points along the side while partly moving in and out through this opening. A larva photographed in the act of hatching and one that had just completed hatching are shown in figure 34. The larva was usually free from the egg shell in less than 24 hours after the first rupture.

Bamboo powder-post beetle was found attacked by a braconid parasite.

While cutting apart a 1-year-old culm of bamboo that had been held in dry storage for about 2 months, a small, brown, wasplike insect was observed in a galley made in the wood by the powder-post beetle (*Dinoderus minutus*). This insect, of the hymenopterous family Braconidae, was tentatively determined as a new species of *Doryctes*.³

The first record of the association of this parasite with the powder-post beetle in bamboo in Puerto Rico was in 1936 by Donald F. Gibbons, who found specimens of the insect at the station in cut bamboo infested by *Dinoderus minutus*. C. F. W. Muesebeck of the Bureau of Entomology and Plant Quarantine, who determined these first specimens, commented that he had seen this form "on two or three other occasions, always recorded as a parasite on beetles, usually on bostrichids in bamboo." Judging from Mr. Muesebeck's later comments, this parasite is common in parts of India and Japan. In Puerto Rico it appears to be relatively scarce and of little importance in the control of this beetle.

CONTROL INVESTIGATIONS OF THE BAMBOO POWDER-POST BEETLE

Previous studies showed good control by copper sulfate introduced into the sap stream of bamboo.

In the 1938 report results were given of an experiment to control the powder-post beetle in 1-year-old *Bambusa vulgaris* culms by means of chemicals applied by a sap-stream method of injection called "stepping." By means of standardized cage tests, it was found that clump curing, zinc meta-arsenite, ammonium bifluoride, and copper sulfate produced outstanding control but were not equally effective throughout the entire length of the culms. The data indicated that this unequal effectiveness may have been caused by unequal distribution of the chemicals and by variations in the starch content of the culms.

Influence of chemical distribution and starch content was verified in a second experiment.

During the winter and spring of 1940, an experiment similar to the foregoing was carried out to clarify the influence of chemical distribution and starch content of the wood on beetle attack. Mercury bichloride at one twenty-fifth the strength of the copper sulfate was used to verify distribution: a 2-percent solution of hydrochloric acid was also employed with the idea of making the wood unattractive to

³ Determined by C. F. W. Muesebeck, Bureau of Entomology and Plant Quarantine.

the powder-post beetle by hydrolyzing the starch in the culm. At the same time, the clump-curing and copper sulfate treatments were repeated for comparison with the results previously obtained, and there was, of course, the usual check or control treatment.

Cage-infestation data were recorded from three parts of the culms.

These treatments were applied separately, as heretofore, to 1-year-old culms in five clumps on the station grounds at Mayaguez. The check or control culms were tested green; the others were harvested for testing about 27 days after treatment. The usual cage-infestation data were recorded, but instead of employing test rings from four internode positions, such rings were taken from three internodes in the culms used. These internodes were (a) the lowest or bottom internode, (b) the one located numerically near the middle of the culm, and (c) the highest one at the top that measured about one-half inch in diameter.

Solutions were absorbed quickly during clear, windy weather.

The strength and amount of the chemicals used is shown in table 28. The dosage applied was at the same rate used in the former experiment, namely, 1 liter of solution per cubic foot of culm volume calculated from the circumference of the base. The average amount of each solution absorbed by the five culms in each treatment is shown also in table 28. This amount was all absorbed by individual culms within about a week, in some cases from 4 to 6 hours after being applied; in general the shortest absorption time was observed to occur, as formerly, during clear, windy weather.

TABLE 28.—*Treatments, concentrations, and amounts of solutions absorbed, and results obtained in an experiment to control the powder-post beetle with chemicals applied by the sap-stream method through the base of Bambusa vulgaris culms; experiment completed May 23, 1940*

Treatment No.	Treatment ¹	Concentration of chemical in solution		Amount of chemical used in 1 liter of solution	Average total volume of culm	Solution absorbed per cubic foot of culm	Beetle attacks—						
							Per 24 rings from—					Total per rings	Control
							Clump V	Clump W	Clump X	Clump Y	Clump Z		
							Number	Number	Number	Number	Number		
1	Untreated check; left uncut until all other culms were harvested, then tested green	Moles	Grams	Cubic feet	Liters		496	434	203	196	169	1,498	0.00
2	Cut off at base and left standing without further treatment				.961		167	131	63	132	88	581	61.22
3	Cut and treated through base with—												
	Commercial copper sulfate	1.3344	333.21		.967	0.991	13	16	4	13	13	59	96.06
4	Mercury bichloride, U. S. P.	.0534	14.50	.890	1.051		34	6	23	116	72	251	83.24
5	Hydrochloric acid, C. P.	.5484	20.00	.841	.933		66	11	169	137	137	520	65.29
	Total						776	598	462	594	479	2,909	----

¹ Applied to separate culms in each of 5 clumps.

Copper sulfate was much superior to clump curing in preventing beetle attack.

In table 28 are summarized by treatments the number of beetle attacks found after 1 month in the rings representing each of the five clumps used and the relative control produced by each treatment. It is evident from the table that copper sulfate produced much better control of the powder-post beetle, 96 percent, than any of the other treatments. Clump curing, which in the first experiment resulted in 91-percent protection, in this test dropped to slightly over 61 percent. Hydrochloric acid gave only 65-percent protection. Mercury bichloride, which was used at one twenty-fifth of the strength of copper sulfate, produced 83-percent control.

According to the usual analysis of variance, the differences between treatments were highly significant, i. e., the odds were 99 to 1 that such differences were brought about by factors other than chance.

Similarly treated culms reacted differently to the powder-post beetle.

It will be noted also in table 28 that there were considerable differences in the number of beetle attacks recorded from the test rings from some of the culms that received the same treatment. While such differences among clumps, as in the previous experiment, were not significant in the majority of cases, the interaction between treatments and clumps was highly significant and pointed to inherent differences in susceptibility to the beetle among culms of the same age in the different clumps. However, where copper sulfate was applied there was no statistical significance in the number of beetle attacks between clumps, indicating that this treatment was equally effective in all five clumps.

The top internode of both treated and untreated culms was attacked less than the middle and bottom.

The number of beetle attacks found in the rings from each of the three internode positions in the culms used is summarized by treatments in table 29.

TABLE 29.—Beetle attacks in test pieces from 3 specified positions in *Bambusa vulgaris* culms treated with chemicals through the base by the sap-stream method; experiment completed May 23, 1940

Treatment No.	Treatment ¹	Beetle attacks—			
		Per 40 rings from internode position ² —			Total per 120 rings
		a, bottom	b, middle	c, top	
		Number	Number	Number	Number
1	None—culm tested green	493	680	325	1,498
2	Cut and cured in clump	427	125	29	581
3	Copper sulfate	25	29	5	59
4	Mercury bichloride	98	102	51	251
5	Dilute hydrochloric acid	195	227	98	520
	Total	1,238	1,163	508	2,909

¹ Applied to separate culms in each of 5 clumps.

² See text p. 79 for explanation.

In table 29 it will be noted that in the experiment as a whole the rings from the top internodes sustained fewer attacks than the rings taken from the middle or bottom internodes. Although the total difference in this respect between the bottom and middle positions was not statistically significant, the differences between the bottom and top and the middle and top were highly significant.

The same trend also occurred within the various treatments, the rings from the top of the treated as well as of the untreated culms being attacked less than the rings from the other two internode positions. In the culms receiving the copper sulfate and mercury bichloride treatments only a few more attacks occurred in the middle internode position than in the bottom where the greatest concentration of those chemicals would be expected. The big difference that occurred in the culms that were clump-cured, where more than three times as many attacks were noted in the bottom as in the middle, was highly significant, as were all the differences among the three positions in the untreated culms that were tested green. In the absence of chemical and physical analyses of the wood itself, these seem to indicate variations, such as in starch or other nutrient content, moisture, or perhaps hardness, in different parts of the culm.

Beetle attack was closely associated with starch content as indicated by iodine test.

That at least some of the foregoing differences might have been brought about by variations in the starch content of the wood in different parts of the culm was again shown by the iodine test. This visual test was applied, as in the previous experiment, to sample rings from the three internode positions in all the culms used. In most of the untreated check culms, the rings from the middle internode were found by this test to contain the highest concentrations of starch, whereas in the treated culms, including those injected with hydrochloric acid, the bottom internode contained the most starch. Except for small differences, which in the case of the chemically treated culms were not significant, these parts were also attacked most by the beetle. Apparently hydrochloric acid under the conditions used had little effect on the starch in these culms.

In two clump-cured culms, beetle attack was closely associated with moisture content.

While this experiment was in progress, an additional sample ring from each position in two of the five culms that had been cured in the clump was set aside for determination of the moisture content. At the time the usual test rings were placed in the cages with the beetles, each sample ring was weighed and then dried to constant weight in an electric oven held at 100° C. The loss in weight, expressed as a percentage of the original weight, was taken as the amount of moisture in the test rings at the time they were exposed to beetle attack. One of the rings from the bottom internode position was thus found to contain as high as 30.9 percent of moisture, while one of those from the top contained as low as 11.1 percent.

While the differences in beetle attack were not quite high enough to be regarded as significant, there was an indication that beetle attack might be linked with the moisture content of the wood at the time of exposure.

Chemical treatments notably affected the wood.

As in the previous experiment, the wood on the inside as well as on the outside of the culms that had been treated with copper sulfate was colored chrome green all the way to the top of the culm. While such a color might not be considered objectionable from some standpoints, the fact that certain metals are corroded in wood impregnated with this chemical may limit the usefulness of bamboo so treated.

Mercury bichloride changed the color of the rind to a light tan. This color was most intense and lasting on the lower half of the culm; the wood on the inside was somewhat darkened. While these changes might not be objectionable from the standpoint of utilization, the poisonous nature of this chemical and its probable corrosive effect on hardware with which it might come in contact would make it undesirable for general use.

Hydrochloric acid faded the color of the outside of the culms, but some of the characteristic green color remained. The greatest change observed was that brought about in the texture of the wood, particularly that in the lower internodes. When sawed into at the end of 3 months after treatment, the wood was found to be brittle, the fibers splitting away to a great extent from the surrounding tissue, and thin pieces of the wood being easily broken. The freshly split wood gave off an odor resembling vinegar which was strongest in the bottom internodes.

Two kinds of powdery excrement were found in borer tunnels.

In considering the effect on beetle attack brought about by copper sulfate and mercury bichloride as used in this experiment, it is of interest to report observations made on the manner in which *Dinoderus minutus* feeds.

Two kinds of powdery excrement were found in the tunnels made by the adults in untreated bamboo wood. When examined under the microscope, one kind appeared as cylindrical to nearly spherical pellets composed mostly of woody material of about the same color as the bamboo, and the other as short, thin fibers or splinters. When a drop of $\frac{1}{40}$ -normal solution of iodine and potassium iodide was placed on these two kinds of excrement the pellets did not change to a blue color, but the splintery material did. This would seem to indicate that the pellets were defecated wood from which all the starch had been extracted by the beetles during digestion and that the splinters, which were as positive to the iodine test as the wood itself, had not been ingested but had been thrown aside while the beetles were making their tunnels.

Beetles were seen to cast aside their borings in attacking bamboo wood.

These observations were confirmed by others made on beetles entering as well as actively boring in bamboo wood, and by subsequent dissection of the same beetles. In starting to enter the wood, the adults were seen to cast aside only splintery, starch-containing material such as that noted above; the only material seen defecated by the insects at this time were small drops of a clear liquid which were soon soaked up by the splinters. When dissected such beetles were found to hold in the intestine only a semiliquid, amber-colored material.

On the other hand, adults actively at work in tunnels already well inside the wood were seen to cast out both kinds of excrement, splinters that were starch-positive and pellets of woody material that were starch-negative. Moreover, beetles in these locations were seen defecating only pellets, and when dissected the intestine was found to contain material of identical form and composition. It is evident that the adult of *Dinoderus minutus*, in common with other members of the family Bostrichidae, may or may not ingest the material it bites loose and thus may avoid, at least to some extent, any poisons it may encounter while boring into the media it infests.

Spot tests indicated that chemical distribution influenced beetle attack and infestation.

Further evidence that some selection such as the foregoing may be exercised by the adults of *Dinoderus* was noted in the test rings from the chemically treated culms. By means of spot tests (5) applied to the two transversely sawed surfaces of the rings from culms that had been injected with mercury bichloride, the bottom internode was found to contain the highest concentration of this chemical, some rings from the middle internode and many from the top not showing a positive reaction. With copper sulfate the reaction was just about as strong in the rings from the top and middle as in those from the bottom.

However, when the spot tests were applied under the microscope, considerable variability was noted in the distribution of the chemicals within the wood itself. Mercury bichloride was located for the most part closely around the fibers and did not extend to the soft, pithy material between the fibrovascular bundles, where the beetle usually enters first and where most of the brood develops. On the other hand, copper sulfate, which could be seen crystallized along some of the vessels, had, as shown by the spot test, not only permeated the fibers but had also saturated most of the spongy pith all the way from the inside of the test ring to the rind.

Few larvae developed in bamboo wood injected with copper sulfate.

During the 3 months following the regular exposure of these test rings to the beetles, many attacks were found and larvae seen to develop in rings from the culms injected with mercury bichloride and very few in those injected with copper sulfate. None of the larvae in the former was found dead, but in the latter dead eggs and larvae, mostly in the first instar, were frequently encountered. In these rings variability in attack and subsequent larval infestation closely followed variability in distribution of the chemical in the wood. At the points of attack and infestation in the mercury bichloride rings, no reaction to the spot test could be seen; at such points in the copper sulfate rings, the reaction to the spot test for copper was hardly any weaker than where no attacks were made. Apparently the relatively lower toxicity of this latter chemical and a slight variation in its intensity in closely connected parts of the wood permitted the few attacks and little feeding that occurred.

Borings in chemically treated culms showed that adults avoided poisoned wood.

As compared with what occurred in the untreated rings, a marked difference was noted in the length of the tunnel and in the character

of the excrement thrown out by the beetles while boring in the rings treated with these two chemicals. In the rings from culms injected with mercury bichloride the tunnels were relatively deep, but the boring was not nearly so extensive as in the untreated rings; in those injected with copper sulfate the few tunnels started were mostly shallow, many less than a millimeter deep. While the character of the excrement thrown out from the tunnels in the mercury bichloride rings was about like that from untreated wood, there was a somewhat larger proportion of fibrous material to pellets. Little of this material and none of the pellets were positive to the spot test for mercury, which indicated that the beetles had cast aside the small amount of poisoned wood encountered. A much larger proportion of fibrous material was found in the excrement from the copper sulfate rings, but most of the fibers and a few of the pellets were positive to the spot test for copper. Since some of the pellets in the excrement showed the presence of copper, it is apparent that some of the poisoned wood must have been ingested. However, it was evident that the beetles were able, probably to an extent inversely proportional to the distribution of the chemical, to avoid the poisoned or lightly poisoned wood.

Uneven distribution of mercury bichloride caused lack of protection from the powder-post beetle.

In a small supplementary test a more thorough distribution of mercury bichloride at the strength used in the foregoing experiment was secured. Sixteen $\frac{3}{4}$ -inch rings from the center internodes of a 1-year-old culm of *Bambusa vulgaris* were dipped in this solution for 10 seconds. When dry, the rings were placed with 50 field-collected beetles in a jar 6 inches in diameter by 9 inches deep. About $21\frac{1}{2}$ months later only 22 attacks, mostly shallow, were found in these rings, only 2 beetles remained alive, the few eggs found were dead, and no larvae had developed. The excrement from the beetles was of the splintery or fibrous type, indicating that little ingestion had taken place. In the same length of time a similar group of untreated rings similarly exposed in another jar were almost completely consumed, and the 50 beetles in this jar had multiplied to such an extent that, on examination, 128 dead and 2,897 living beetles were found.

Since many larvae developed in the rings from culms that had been injected with mercury bichloride solution and none in the rings that had been dipped in a solution of the same strength as described above, it is apparent that lack of protection in the injected culms was caused by uneven distribution.

Though less toxic, superior distribution of copper sulfate afforded better protection than mercury bichloride.

No supplementary tests, such as that just described, were made with the copper sulfate solution. However, much evidence of the action of this chemical when injected into the sap stream of bamboo culms was gained from the examination of the rings here reported. From the positive results of the spot tests of the wood and of the beetle excrement, and the fact that some larvae did at least partially develop in the test pieces, it may be concluded that copper sulfate as here used was not sufficiently toxic to cause immediate death of the

beetles or the larvae, but that attack and larval development were held at a minimum by the relatively thorough distribution of the chemical in the wood.

Entomological investigations were carried on by Harold K. Plank, associate entomologist.

CHEMISTRY INVESTIGATIONS

Excellent wine can be made from juice of Puerto Rican oranges.

In the annual report for 1938 it was mentioned that three types of palatable wines could be made from the juice of oranges provided the amount of sugar added was controlled so that, even though the same percentage of alcohol was obtained in each type, the amount of unfermented sugar remaining in the wines varied from a low to a high percentage. In this study, orange juices were obtained that varied considerably in Brix reading and in citric acid and total sugar content. The fruits from which these juices were obtained were bought in the local market and might have been mixtures of varieties or in varying degrees of maturity.

While a higher sugar content of fresh orange juice means a lower requirement in the addition of sugar to obtain the necessary alcoholic content in the fermented product for good keeping qualities, it is also true that the higher the citric acid content of the fermented product, the higher the sugar requirement to make it palatable. Thus, while the orange season in Puerto Rico ordinarily runs from November to April or May, there is a possibility that the fruits obtained in some of these months may be low in sugar and high in acid or otherwise not suitable for a first-class wine.

The influence of maturity on the amount of juice, acid, and sugars in local oranges was studied.

A study was therefore made of the juice from oranges in varying degrees of maturity throughout the picking season, with a view to determining the maturity which might be expected to yield the best juice for wine making. The oranges used were picked at random from two trees on the same farm, but at different dates and hence different stages of maturity. At each harvest date the fruits representing extremes in maturity for the particular harvest date involved were discarded. The fruits picked for the first analyses, about the middle of November, were fully developed oranges but with a tough, dark-green rind with no sign of ripening. Analyses were made of the juice of fruits from the same trees subsequently picked approximately every 15 days until the end of April, at which time the fruits started to show signs of rind decay. The next day after picking the fruits were peeled and the juice extracted with a hand press. At each harvest records were made of the appearance of the fruit, the juice extracted in terms of gallons per 100 pounds of fruit, the pH and Brix of the juice, and the reducing sugars, sucrose, and total sugar content of the juice. The results of these periodical analyses are given in table 30.

TABLE 30.—*Analyses of juice from representative oranges picked from 2 trees at Mayaguez, P. R., at approximately 15-day intervals from Nov. 17, 1939, to Apr. 30, 1940*

Sample No.	Date harvested	Color, maturity, etc.	Extraction of juice per 100 pounds fruit ²	Analysis of juice ¹					
				Soluble solids	Citric acid	Acidity	Sugars		
							Reducing	Sucrose	Total
	1939		Gallons	Brix	Percent	pH	Percent	Percent	Percent
1	Nov. 17	Deep green, immature ³	4.99	8.9	1.81	3.01	3.14	2.31	5.45
2	Dec. 2	Light green, immature ³	5.23	9.4	1.91	2.96	3.29	2.71	6.00
3	Dec. 31	Light green, slightly mature ⁴	5.42	10.1	1.71	3.08	3.53	3.10	6.63
	1940								
4	Jan. 15	Light green spotted with light orange, mature ⁵	5.28	11.0	1.50	3.17	3.88	3.72	7.60
5	Jan. 30	Light orange, ripening ⁵	5.73	11.5	1.40	3.28	4.09	4.04	8.13
6	Feb. 14	Orange, ripe ⁵	6.08	11.8	1.20	3.42	4.02	4.42	8.44
7	Mar. 2	do. ⁵	6.42	11.9	1.20	3.45	4.11	4.45	8.56
8	Mar. 18	do. ⁵	6.66	12.0	1.12	3.58	4.20	4.40	8.60
9	Mar. 31	do. ⁵	6.60	11.9	1.05	3.67	4.10	4.40	8.50
10	Apr. 15	Orange, wrinkled, overripe	6.42	11.4	.98	3.71	4.09	3.93	8.02
11	Apr. 30	Orange, wrinkled, a few fruits beginning to decay	6.47	10.8	.92	3.76	3.83	3.73	7.56

¹ Each value represents the average of 2 determinations.² Based on juice extracted from 15 fruits.³ Fruits firm, too hard in texture; juice extracted only with considerable pressure.⁴ Fruit texture somewhat softer than that of sample No. 2; juice extracted with less pressure.⁵ Texture soft, yielding when squeezed in palm of hand; juice extracted with maximum ease.

It can be seen from table 30 that, beginning about the middle of November with distinctly immature oranges, the quantity of juice extracted per 100 pounds of fruit, the total soluble solids in the juice as indicated by Brix readings, and the percentage of reducing sugars, sucrose, and total sugars in the juice increased steadily, with only two exceptions, until the fruits became fully ripe in March; thereafter as the fruits became progressively riper and eventually began to break down, the juice qualities retrogressed. Both the citric acid content of the juice and the concentration of hydrogen ions in the juice as measured by the pH decreased consistently, with one minor exception, through the entire period during which the fruits were sampled.

These results indicate that oranges as ordinarily placed on the market in Puerto Rico during November and the early part of December are not in the most desirable condition for the manufacture of wine. During this period the fruits were hard and much pressure was necessary to press out the juice and the quantity of juice obtained per 100 pounds of fruit was low. The juice qualities from about the latter part of February through the middle of March were at their peak for the economical manufacture of high-quality wine. Desirable qualities in the juice were correlated with the presence and depth of the orange color of the rind and a general lack of firmness as evidenced by the fruits yielding easily to pressure exerted in the palm of the hand.

Oranges are produced in Puerto Rico from near sea level up to an altitude of approximately 3,000 feet. In general, fruit produced at higher elevations tends to mature later than that grown near sea level. As a consequence, fruits of varying degrees of maturity may be found on the market at any one time. However, by February most

of the fruits on the market are sufficiently mature to be of acceptable edible quality and to make palatable wine without the addition of large quantities of sugar. Aside from the low yield of juice, low sugar and high citric acid content of oranges available early in the season,

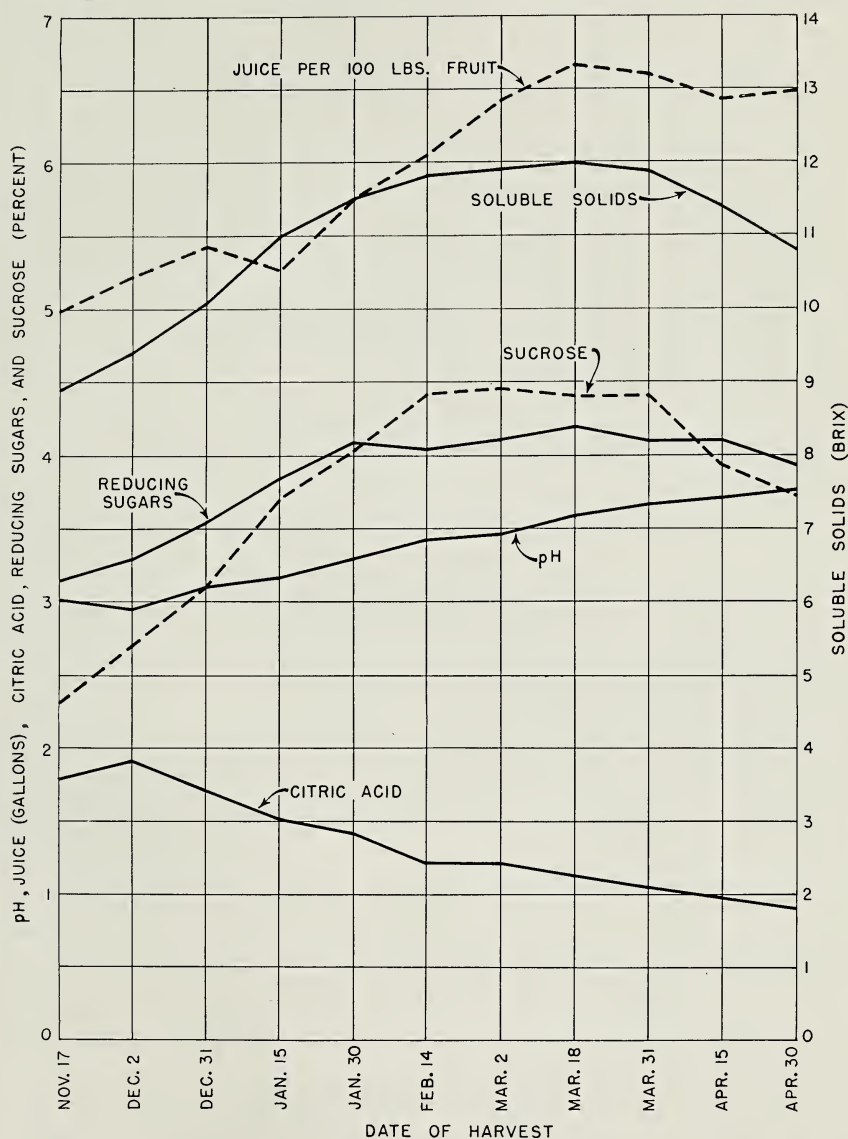


FIGURE 35.—Physical and chemical changes associated with maturity in Puerto Rican oranges as indicated by gallons of juice per 100 pounds of fruit, Brix and pH of the juice, and citric acid, reducing sugars, and sucrose content of the juice.

the flavor of the finished product is adversely affected by using fruits picked before they are fully ripe.

The progressive changes during maturity in gallons of juice per 100 pounds of fruit, Brix and pH of the juice, and citric acid, reducing

sugars, and sucrose content of the juice are shown graphically in figure 35.

Cooperation was extended to industry and to other agencies of the Federal and Insular Governments.

The station cooperated on several occasions with persons interested in the manufacture of cane sirup and orange wines. On three occasions sirup plants were visited and methods suggested which remedied failures to obtain a good manufactured product. On two occasions persons who were greatly interested in the manufacture of orange wines were given a detailed description of the best methods to use to obtain the most palatable product, and of the apparatus required for such manufacture.

Cooperation was also given to other divisions of the experiment station and some agencies of the Federal and Insular Governments whenever this was required. Especial attention was given to cooperation in the coffee-nutrition studies being carried on in one of the station greenhouses by the Agricultural Experiment Station of the University of Puerto Rico.

Some attention was given to analysis of fertilizers, minerals, deposits from caves, and other materials the examination of which was considered important. Waters for drinking and irrigation purposes were also examined.

The foregoing investigations were conducted by José O. Carrero, assistant chemist.

The nonvolatile acids and flavor of the soursop were studied.

The fruit of *Annona muricata* L., known in the Spanish West Indies as guanábana, or the soursop, is large and fleshy, often as large as a child's head and weighing as much as 5 pounds. It is ovoid or heart-shaped, dark green, the glabrous skin bearing numerous recurved fleshy spines, and its pulp is white and juicy, pleasantly subacid, with a pleasing flavor. It is grown with especial excellence in Puerto Rico. A favorite drink is made from the juice, and the pulp yields excellent jelly, tarts, and preserves. The tree which bears this fruit is about the size of a peach tree and is in general cultivation throughout the Tropics of both hemispheres.

Analysis of this fruit has been made by Stahl (12, p. 13) and proximate analyses are reported by Wehmer (17, p. 592). Analyses of *Annona muricata* fruit grown in Cuba were made by Chace, Tolman, and Munson (6, p. 23); of Philippine fruit, by Pratt and Del Rosario (11, p. 76); and of Hawaiian fruit, by Thompson (13, p. 67). These references are given by Winton and Winton (18, p. 529). However, no investigation of the nature of the flavor appears to have been made previous to the present study.

The fruit was analyzed and its vitamin C content determined.

Soursop fruits from the station grounds at Mayaguez were found to have the following analysis:⁴

	Percent
Total solids.....	16.72
Soluble solids.....	14.57
Insoluble solids.....	2.15
Ash.....	.71
Alcohol precipitate (pectin).....	.91

⁴As determined by A. L. Curl, Food Research Division, Bureau of Agricultural Chemistry and Engineering.

	Percent
Total sugars (as invert)-----	11. 19
Acidity (milliliters 0.1-normal NaOH per 100 grams)-----	84. 5
Vitamin C per milliliter juice (milligrams)-----	. 08

Vitamin C was determined by titration with 2,6-dichlorophenol-indophenol.

The acids consisted of a mixture of malic, citric, and isocitric acids.

The juice from 21 kilograms of peeled guanábanas was extracted by boiling the pulped fruit and pressing out the extract in a hydraulic press. The extract was diluted with an equal volume of alcohol and the precipitate (pectin, etc.) was removed by filtration. The filtrate was then neutralized with sodium hydroxide, and the organic acids were precipitated as lead salts with a solution of lead acetate.

The acids were recovered from the lead salts by adding sulfuric acid, and the slight excess of sulfuric acid was removed from the filtrate from lead sulfate with just sufficient barium hydroxide.

The acid filtrate was then evaporated to dryness and esterified by refluxing with absolute ethyl alcohol containing 2.5 percent hydrochloric acid.

The esters were extracted from the residue left after evaporating the alcohol by solution in ether; the ether solution was filtered and washed with a dilute solution of sodium hydroxide and the ether was evaporated. The 50 grams of ethyl esters were then fractionally distilled through a 15-centimeter Widmer column at 10 millimeters, yielding the following fractions:

Fraction 1, 122–127° C.	= 31. 72 grams
Fraction 2, 170–172° C.	= 16. 26 grams
Flask residue	= . 7 gram

Hydrazides were prepared from these fractions. Fraction 1 yielded a hydrazide melting at 177.5–178.5° C. It gave no depression in melting point when mixed with *l*-malic hydrazide, and was therefore presumed to be identical with that compound. Fraction 2 gave a hydrazide melting at 105–106°. It gave no depression when mixed with citric hydrazide, indicating the two were identical; it was also identified as citric hydrazide by optical crystallographic examination.⁵ From the flash residue a small amount was distilled from a microflask which afforded a hydrazide melting at 173–174°; this gave no depression when mixed with isocitric hydrazide, and was identified as such by optical crystallographic examination.

The acids of guanábana therefore consist of a mixture of about two parts of malic acid, one part of citric acid, and a trace of isocitric acid. In determining flavor, light alcohols and acetaldehyde were separated.

Forty-two kilograms of guanábanas were peeled, pulped, and distilled with steam. The distillate was redistilled several times to smaller volume each time and the final distillate, amounting to 600 milliliters was distilled through a 15-centimeter Widmer column.

The following fractions were separated:

1-----	at 60–80° C. (aldehyde odor).
2-----	at 80–100° C.
3-----	at 100° C. (turbid, odor of fruit).

Fractions 1 and 2 were saturated with salt, extracted with ether and the ether was added to an ether extract of fraction 3.

⁵ Made by G. L. Keenan, Food and Drug Administration.

The salt solutions of fractions 1 and 2, after ether extraction, were again distilled through a Widmer column, yielding:

Fraction 1	-----	under 75° C.
Fraction 2	-----	at 75-79° C.
Fraction 3	-----	at 79-81° C.

The total amount boiling under 81° was 15 milliliters. The material afforded a strong aldehyde test with Schiff's reagent. When it was treated with semicarbazide hydrochloride and sodium acetate solution, and the alcohols were removed by distillation, a semicarbazone was obtained, which melted at 161-162° and gave no depression in melting point when mixed with acetaldehyde semicarbazone.

The alcohols recovered from the semicarbazone separation afforded a strong reaction for methyl alcohol by the U. S. P. test (*16, p. 355*). On careful repeated refractionation through a Vigreux column, a fraction boiling at 77-79° was obtained which was oxidized with potassium dichromate mixture and distilled. The distillate had the odor of acetaldehyde and gave a deep pink color with Schiff's reagent. The unoxidized alcohol gave only a slight color with this reagent.

The light-boiling fraction therefore appears to be a mixture of methyl and ethyl alcohols and acetaldehyde.

Flavor was also found to be influenced by an oil composed of several esters.

On evaporation of the ether from the ether extracts of the distillate from guanábana fruits, 1.58 grams of an oil was obtained. This possessed the characteristic odor of guanábana in concentrated form.

A drop of the oil evaporated on a strip of paper gave a first impression of an amyl ester. The residual odor resembled geraniol, or rather a geranyl ester.

The oil was saponified and the alcohols were distilled with steam and recovered from the distillate by ether. The residue left on allowing the ether to evaporate spontaneously had the cough-producing property and odor of amyl alcohol. A drop evaporated on a strip of filter paper gave first the odor of amyl alcohol, then an odor resembling one of the higher aliphatic alcohols such as octyl alcohol, and finally a pronounced odor of geraniol. When the alcohol mixture was distilled in a microstill and the fractions were examined separately, the first fraction, oxidized with bichromate mixture, gave the odor of valeric acid; an odor of citral was obtained on oxidation of the higher fractions.

Various caproates contribute to guanábana flavor.

The alkaline solution obtained on saponifying the flavor oil was acidified with phosphoric acid, diluted with water to 30 milliliters, and distilled.

The first fraction, about 15 milliliters, was kept separate. It contained undissolved droplets of oil. These were separated by filtration through a wet filter.

The undissolved acid required 5.4 milliliters of 0.1-normal NaOH to neutralize it, and the acid in solution required 18.55 milliliters.

Silver salts were prepared from each. The proportions of silver in them were:

	Percent
In undissolved-acid salt	Ag=48.40
In dissolved-acid salt	Ag=49.04
Calculated for silver caproate	Ag=48.38

The acid would therefore appear to be mainly caproic acid, and the odor of the acid confirms this conclusion.

Concentration of the filtrate from the silver salt, and examination of a second fraction obtained on distilling the acids, which required only 1.9 milliliters of 0.1-normal NaOH, failed to reveal the presence of any acetic acid.

Mixtures of amyl caproate, geranyl caproate, and small amounts of amyl butyrate and acetaldehyde resemble the guanábana flavor, but there is something lacking. The natural flavor has a smooth note, the nature of which was not discovered.

From the studies here reported it can be concluded that the organic acids of the guanábana or soursop consist of a mixture of malic and citric acids in approximately the proportion 2:1. A trace of isocitric acid was also found.

The flavor of guanábana is evidently due, at least mainly, to a mixture of amyl caproate with geranyl caproate, and possibly other caproates.

The nonvolatile acids of the carambola were investigated.

The fruit of *Averrhoa carambola* L., commonly known as carambola, is ovate, acutely five-angled, star shaped in cross section, with a yellow, waxy skin and an acid pulp. Specimens examined varied from 41 to 122 grams in weight. The seeds average about eight per fruit, each seed averaging about 0.05 gram, and are similar in appearance to apple seeds. The fruit is said to be used for pickles when in the half-grown state and for preserves when ripe, and is also said to be used extensively by the Chinese in various fish dishes.

The carambola tree is cultivated in India and China, sparingly in southern California, and frequently in the West Indies, and it also occurs in Hawaii, Guam, and the Philippine Islands.

Proximate analyses of the fruit are given by Wehmer (17, p. 592), but no mention is made of the acidity. Winton and Winton (18, pp. 680, 681) report analyses by Pratt and Del Rosario (11, p. 76) and Thompson (13, p. 67) and call attention to an evident jumbling of the figures for the sweet and sour varieties. Pratt and Del Rosario were the first to call attention to the presence of oxalic acid. A later analysis was made by Stahl (12, p. 13), but he did not mention the nature of the organic acids present, which was the chief object of the work here reported.

Ripe fruits of the sour variety grown in Puerto Rico contained much acid.

A quantity of ripe fruits from several trees of the sour variety, introduced by and planted at the station in 1923, was analyzed, with the following results:

	Percent
Total solids.....	7.49
Soluble solids.....	6.27
Insoluble solids.....	1.22
Ash.....	.28
Alcohol precipitate (pectin).....	.53
Total sugars (as invert).....	4.86
Acidity (milliliters 0.1-normal NaOH per 100 grams).....	144.8
Vitamin C per 1 milliliter juice (milligrams).....	.11

Vitamin C was determined by titration with 2,6-dichlorophenol-indophenol. Analysis of the 0.28 percent ash shown above gave:

	Percent
Fe ₂ O ₃ +Al ₂ O ₃ -----	0.005.
CaO-----	None.
MgO-----	0.013.
K ₂ O-----	0.14.
Na ₂ O-----	Not determined.

Traces of malic acid and citric acids were identified.

The juice from 8.5 kilograms of fruit was diluted with alcohol, neutralized with sodium hydroxide, and precipitated with lead acetate.

The acids recovered from the lead precipitate were crystalline. They were esterified by refluxing with ethyl alcohol containing 2.5 percent of hydrochloric acid. The ether solution of the esters, after filtering and washing with sodium hydroxide solution to remove excess acid, was evaporated on a steam bath.

Most of the ester evaporated at rather low temperature, which pointed to the presence of a low-boiling ester, such as ethyl oxalate.

From the residue left on evaporation, ether extracted a very small amount of ester, and this, treated in alcoholic solution, gave a precipitate of hydrazide which melted at 179° C. after recrystallization, and on admixture with *l*-malic hydrazide gave no depression.

From the filtrate from malic hydrazide another hydrazide was precipitated more slowly. This had the appearance of citric hydrazide and its identity with that hydrazide was established by optical crystallographic examination.⁶

Acidity was found to be due to oxalic acid, mostly in the free state.

A direct examination for oxalic acid was made by the Arbenz (1) method. Five hundred and fifty-six grams of fruit, expressed in a Carver press, yielded 410 milliliters of juice. Ten milliliters of juice required 13.2 milliliters of 0.1-normal NaOH, equivalent to 0.594 gram of acid, calculated as oxalic, per 100 milliliters.

For the determination of total oxalic acid, 100 milliliters of juice were concentrated to 20 milliliters strongly acidified with hydrochloric acid, and extracted for 24 hours in an apparatus designed for continuously extracting liquids with ether.

The residue from the ether was dissolved in dilute ammonia, made acid with acetic acid and precipitated with calcium chloride solution. The precipitate was filtered, redissolved in hydrochloric acid, made alkaline with ammonia, then acid with acetic acid, and the calcium oxalate reprecipitated. Ignition of the precipitate yielded 0.311 gram CaO, equivalent to 0.5 gram oxalic acid per 100 milliliters juice.

Repetition of the operation on 100 milliliters of the unacidified juice afforded 0.41 gram of free oxalic acid per 100 milliliters of juice, leaving 0.09 gram of combined oxalic acid, which is probably in the form of acid potassium oxalate, as potash constitutes one-half of the total ash.

The juice of the carambola, when mixed with water and sweetened, makes a pleasant acid drink. However, because its acidity is due to oxalic acid, carambola juice cannot be compared to lemon juice but

* Made by G. L. Keenan, Food and Drug Administration.

rather to rhubarb juice, which also contains oxalic acid. This may be of interest to the physician in cases where the ingestion of oxalic acid or oxalates is undesirable.

It is evident from this investigation that the predominating organic acid in ripe carambola fruits is oxalic acid, 0.5 gram per 100 milliliters of juice, and that this is mostly in the free state. Traces of malic and citric acids are also present. The vitamin C content, 0.11 milligram per milliliter of juice, is not high.

These investigations of the nonvolatile acids of the soursop and carambola were the work of E. K. Nelson, senior chemist, now deceased, and A. L. Curl, assistant chemist, of the Bureau of Agricultural Chemistry and Engineering, Washington, D. C.

AGRICULTURAL ENGINEERING

Ditcher was developed for use on sugarcane land.

Under the heavy rainfall conditions in parts of Puerto Rico, drainage is often important, especially in level fields. Since sugarcane occupies more of the level or nearly level land than is occupied by all other crops combined, the need for drainage is felt most keenly by cane growers.

Machines for ditching are being built by implement manufacturers, but it is felt that ditches made with these machines are too wide for use as laterals. Cane growers have indicated that they would like to have these lateral ditches approximately 20 inches deep and as narrow as is feasible to build them. One manufacturer's specifications show that a ditch 20 inches deep is 14 inches wide at the bottom and 48 inches wide at the top. Ditches this size spaced 40 feet apart will occupy 10 percent of the area of the field. Thus cane growers operating on land valued at from \$500 to \$800 per acre rightfully feel that too much investment is set aside in ditches when the ditches are as much as 4 feet wide.

First model worked satisfactorily in friable soils.

From a design prepared by the station, and under its general supervision, an implement was built by one of the plantations to make ditches 20 inches deep, 6 inches wide at the bottom, and 24 inches wide at the top. Insufficient time has elapsed since the construction of the ditches made with this machine to determine whether the earth from the steep bank slopes will slide to the extent of blocking channels and thus interfere with drainage. The ditches made with this machine are shown in figure 36.

This ditcher performed fairly satisfactorily in friable soils in spite of the fact that it was crudely built from boiler plate in a blacksmith shop. Considerable hand labor was required to complete the ditch, but this was only a fraction of that required to dig the whole ditch by hand or to plow and complete by hand. Its performance on more difficult soils was unsatisfactory, but even in the stiffest soils a large amount of labor was saved.

In constructing the ditcher, two wings, made from boiler plate following the general shape of the moldboards of a Cotton States middle-buster, were mounted on a 36-inch standard Killefer subsoiler. Long additional wings of the same material were provided on the upper parts of the moldboards to spread the soil 5 feet on either side of the



FIGURE 36.—Ditches made with experimental ditcher without aid of any manual clean-up work.



FIGURE 37.—Battery of three stills showing the condenser tank at the left. The center retort has the cap removed and placed on a table provided for this purpose. The basket is partly removed and is shown suspended on the hoist.

ditch. Angle iron braces and reinforcements were used to secure sufficient strength. It was pulled by a 50-horsepower track-type tractor.

Judging from the results secured from this crude implement, it seems reasonable to assume that a ditcher can be built along these general lines that will prove entirely satisfactory in the building of laterals on most soil types on which cane is grown on the island.

Experimental essential-oil stills were completed during September.

A battery of three experimental stills for the recovery of essential oils from aromatic plants was completed during September. This battery is shown in figure 37. The boiler for the generation of steam was one being employed for other purposes. Stills were required that would be adaptable for use with a number of kinds of plants the flowers, leaves, or fruits of which yield oil. It was, therefore, impor-

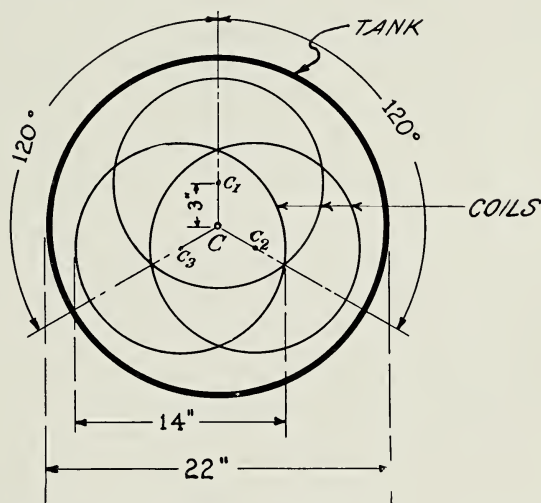


FIGURE 38.—Arrangement of coils in condenser tank; C , Center of tank; C_1 , C_2 , and C_3 , centers of coil units, spaced 3 inches from center of tank.

tant that these stills could be successfully operated under various conditions, such as with live steam, with hot water, and at reduced pressures and temperatures. To meet certain experimental conditions, an accurate control of the pressure was desirable, and it was important that provision be made for measuring both the pressure and the temperature. The use of metal that would be inactive in contact with steam and the various essential oils was also essential throughout the construction.

The stills were designed and the specifications written to meet all of these conditions with slight modifications, and the retorts were manufactured under contract according to these specifications.

Each still is a cylindrical body 14 inches in diameter and 19 inches high, with conical top and bottom. This accommodates a basket 13.6 inches in diameter and 18.5 inches high with a capacity of 10 gallons dry measure. The retorts are made of heavily tinned copper, and equipped with gate valves top and bottom to control steam pressure,

a safety valve, drain plug, thermometer well, and pressure gage. Each retort is also drilled for the installation of a glass sight gage and for the circulation of water, if such should be found desirable.

Three interlacing coils, set in a common cooling tank, serve as condensers. These coils, made of pure tin tubing three-eighths of an inch in diameter, were constructed at the station, extreme care being taken to make them uniform so that the condensing surface would be identical in each. A single cooling tank was used in order to insure a uniform condensing temperature for all units. The tin tubing was bent into three interlacing coils having $8\frac{1}{2}$ turns each 14 inches in diameter. These coils were then inserted into a tank 22 inches in diameter and 18 inches deep, as shown in figure 38. The end of the pipe that leads cooling water into the bottom of the tank was so bent as to give a swirling motion to the water to aid in securing uniform water temperatures on all three coils.

It was calculated that each condenser coil would condense the steam required to recover the oil from 10 gallons of material in 1 hour with a temperature rise of 40° C. in the cooling water. The data on the amount of steam required in this calculation were secured from the operation of a small laboratory still during the recovery of oil from lemon grass. The volume of steam required will doubtless vary under different conditions and with different plant materials.

These stills have been used to recover oil from a number of plants and their operation has been completely satisfactory.

Experimental solar drier effectively reduced moisture content of bamboo wood.

In certain bamboo work there is need for material containing less moisture than can be obtained by air-drying in the atmosphere at Mayaguez. A small wooden box was constructed for drying strips of bamboo in the sun. This drier is 6 inches wide, 6 inches deep, and 8 feet long. The box was covered with black metal on the outside, and small holes were drilled in bottom and top for limited air circulation. When exposed to the full force of the sun's rays for a few hours, the inside temperature was observed to rise 25° C. above that outside, the maximum temperature observed being 55° . The moisture content of samples of bamboo kept in this box for 5 days was reduced from an original of 11 percent to a final 5 percent on a dry-weight basis. This drying box, though effective during the dry season, proved to be of little value during the rainy season, during which drying tests similar to the above but extended over a 10-day period removed only a negligible amount of moisture from the bamboo samples. It is assumed that the few hours of effective sun during the rainy season is insufficient to effect a worth-while amount of moisture removal.

James K. Alvis, assistant agricultural engineer, has been in charge of agricultural engineering activities at the experiment station during the year.

IMPROVEMENTS IN PROPERTY

Upper Jagua dam was completed.

An earth dam in which the core was puddled with sodium chloride to obtain maximum agglutination of soil particles was constructed in

the upper reaches of the Jagua valley. The dam has a capacity of $3\frac{1}{2}$ acre-feet and the spillway is 24 feet above the floor with a 4-foot freeboard. The dam has three outlets similar to the one shown in the 1938 annual report and not only functions for holding irrigation water during the dry season, but, with the middle outlet open during the wet season, also gives control over floodwaters within its capacity in periods of excessive rains.

This is the fourth earth dam of this nature constructed at the experiment station, and apparently is one of the best investments of human labor possible to convert the liabilities of the excessive rains and steep-sided valleys of the island into assets for the community.

Terraces were constructed for plant introductions.

In the lower Jagua valley to the north of reservoir No. 1, one-third of an acre of wide bench terraces was constructed upon which to extend the constantly expanding collection of introduced tropical plants of the experiment station. These bench terraces, although too expensive for commercial practice, can be adapted so well to the careful charting of locations of valuable plant introductions that some unusual expenditure in their construction is justified. The terraces are well situated, not only serving as an extension of the botanical garden, but also contributing to the landscaping of the station as a whole.

Utility house was constructed at Maricao.

In the municipality of Maricao, 20 miles distant, the experiment station has an area of 30 acres contributed by the Insular forest service. This area of land, at an elevation close to 3,000 feet above sea level, serves for acclimation studies of new crops, and particularly for investigations of crops requiring high elevations, such as quinine and some of the other alkaloid-producing crops. During the year a utility house with concrete foundations and wooden superstructure was constructed to serve for tool storage and to provide laboratory and work space for research workers. The house, with floor space of 860 square feet, is also provided with a room and bunks for the housing of members of the staff for short periods. A 2,000-cubic-foot cistern of reinforced concrete was built under the house as a source of potable water supply. Labor and materials for this house were furnished by the Puerto Rico Reconstruction Administration; construction was under the supervision of the experiment station technical staff.

Second staff residence on Las Mesas was completed.

Using materials from one of the barracks vacated by a camp of the Puerto Rico Reconstruction Administration, a second staff residence at Las Mesas was completed in April 1940. The house is located at an elevation of 1,000 feet above sea level, with a splendid view of the Añasco valley on the north, Mona Passage on the west, and the San German Valley and Carribean Sea on the south, contributing to the pleasantness of such living quarters. The house, of wooden construction on concrete foundations, to avoid termite damage, has a total floor space of 2,000 square feet.

A valuable development, to some extent also experimental, was the construction under the house of a concrete cistern to store rain water

for household purposes. Las Mesas during the dry season has an acute water shortage, which has limited to a considerable extent the use of the extensive land area there for residential purposes. However, during the rainy season there is an average annual rainfall of some 90 to 100 inches. This cistern, with a capacity of 48,000 gallons, filters the rain water from the roof and stores it for the dry season, and an automatic electric pump delivers the water under pressure from the cistern to the house above. The residence has contributed greatly to relieve the housing shortage for members of the technical staff.

Shelf space of library was trebled.

The library of the experiment station is not extensive, but is apparently complete for the publications of the Department of Agriculture, while those of the agricultural experiment stations of the United States as well as agricultural institutions throughout the Tropics of the world are well represented. The library has never been adequately housed. To remedy this partially, new shelving was constructed throughout the library, reaching from floor to ceiling, which has trebled the shelving capacity. Additional space for the library is still needed.

Plans for new laboratory wing to the main station building were completed.

During the year a project was approved by the Work Projects Administration for the construction of a new laboratory wing to the main laboratory and office building of the station. Plans were drawn up and approved which provide 8 new laboratories with a total floor space of 7,500 square feet. Four of these laboratories will be especially equipped for chemistry investigations. The building also provides for more modern wash and rest rooms, a great need of the station at the present time. The building will be of reinforced concrete with tile floors, fluorescent lighting, copper tubing for delivery of water, gas, and compressed air, and as permanent and effectively arranged as modern design will permit.

The flat tiled roof of the building will be accessible by an outside staircase, making it available for open-air assemblies, which are practical in the comparatively regular climate of Mayaguez. All laboratories will have a pleasant outlook and circulation of air, contributing an unusually favorable environment for research. This new laboratory wing will increase the facilities of the experiment station in its function as a research outpost in the Tropics for the Department of Agriculture.

General upkeep of station buildings is extensive.

In the moist, warm climate of Mayaguez, deterioration of wooden buildings is comparatively rapid. Heavy rains also make necessary constant attention to roadways, bridges, culverts, and fields to secure adequate and rapid distribution of excess water. During the year extensive repairs were made to wooden buildings, substituting concrete construction wherever possible. Roadways have been improved. The electric lines have been relocated to avoid marring landscaping effects. Two laboratories were remodeled to make them available for chemistry investigations. Considerable new laboratory furniture was built.

Improvements in property have been under the supervision of James K. Alvis, assistant agricultural engineer, and Pedro Folch, junior agricultural engineer.

COOPERATION WITH OTHER GOVERNMENT ORGANIZATIONS

The P. R. R. A. materially advanced station objectives.

During the year 3 technical men were employed by the Puerto Rico Reconstruction Administration to aid at the station in investigations in the production and processing of essential oils, the production of insecticidal crops, and the production and processing of vanilla. Relief labor to the extent of 60 men was also provided to aid in the foregoing investigations, as well as in the studies of drug plants, spices, and insect-resistant bamboos. The investigational results reported in the foregoing pages on those subjects have been greatly aided by the contributions and cooperation of the P. R. R. A. A total of \$26,000 was budgeted by the Reconstruction Administration for these activities during the year.

Civilian Conservation Corps administered by Forest Service has been invaluable.

During the year from 50 to 60 men were made available by the C. C. C. for the propagation of the newly introduced insect-resistant industrial bamboo species, the new mango varieties which have so much consumer demand, and the propagation of quinine and other drug crops. The labor and supervisory personnel of the C. C. C. have been outstanding for their effectiveness and accomplishment.

New projects were undertaken by National Youth Administration.

During the latter part of the fiscal year personnel was contributed by the National Youth Administration for the dual purpose of aiding in the objectives of the experiment station and contributing training to such personnel. Young men and women were made available for library work, assistants in the chemical laboratories, assistants in the field work of the station, and in the designing and construction of articles in the bamboo shop. The personnel assigned to the bamboo shop, in particular, has shown great interest in such work, often returning outside of the usual work hours to continue their activities. Appreciation is also expressed to the administrative officers of the N. Y. A. for their complete cooperation in furthering the objectives of the experiment station.

Cooperation of Government of Puerto Rico has greatly advanced research projects.

As in recent years, the Government of Puerto Rico has contributed funds for the employment of technical personnel in the production and processing of vanilla, the production of spice and other tropical crops, studies of the production and processing of essential oils, and the production and utilization of bamboo. This personnel has not only contributed technical knowledge of value to Puerto Rico, but some of the contributions are of definite value to the economy of the whole United States. Cooperation has continued and become more intimate with the Insular Department of Agriculture and Commerce, the College of Agriculture and Mechanic Arts,

and the Experiment Station of the University of Puerto Rico. Appreciation is expressed to the administrative officers and personnel of these entities for their constant attitude of cooperation.

Experiment station houses other Federal activities.

Extensive office and laboratory space, as well as field areas, have continued to be made available to the Soil Conservation Service and the Bureau of Entomology and Plant Quarantine of the Department. A plant-quarantine inspector of the Insular Government has also been given office space at the experiment station. During the latter part of the year office space was made available to clerical officers of the Puerto Rico Reconstruction Administration in order to economize on their office rentals in the city of Mayaguez.

Correlating Committee has filled essential role.

The experiment station has maintained constant attendance at meetings of the Correlating Committee. This committee consists of representatives of the Puerto Rico Reconstruction Administration, the College of Agriculture and Mechanic Arts, the Experiment Station, and the Extension Service of the University of Puerto Rico, the Department of Agriculture and Commerce of Puerto Rico, the Division of Vocational Training of the Department of Education, the United States Department of Agriculture Soil Conservation and Forest Services, and this experiment station. The committee has seemed to have a highly important function in utilizing to the best advantage the resources and appropriations made available for agricultural development in Puerto Rico.

PUBLICATIONS

Monthly Report series included 90 individual staff reports.

As in previous years, the regular divisional monthly reports, showing the development of the projects of the station, were issued in mimeographed form for interoffice circulation. There were 12 issues in this series, 1 for each month, and 6 supplements to the June issue, in all covering the subject matter of 90 individual reports, totaling 286 pages.

Besides being circulated among the personnel of the Department, copies were also frequently sent on request to planters and others maintaining a close professional interest in the activities of the station. The mailing list included 40 names.

Nine papers were published by station workers.

Several papers, reflecting some of the results of work carried on at the station, were prepared by members of the staff. The following bibliographical list contains eight such articles published during the year under review and a previously unreported article that appeared during the fiscal year 1938.

- BAILEY, WALLACE K. Summary of results of experiments in controlling corn ear pests in Puerto Rico. Amer. Soc. Hort. Sci. Proc. (1939) 37: 763-769, illus. 1940.
- BARTLETT, K. A. The results of shipments of the predatory elaterid beetle *Pyrophorus luminosus* from Puerto Rico to England. Bul. Ent. Res. 30: 209-210. 1939.

- BARTLETT, KENNETH A. The collection of parasites of the sugarcane borer, *Diatraea saccharalis*, in São Paulo, Brazil. Sixth Pacific Sci. Cong. held July 1939.
- BARTLETT, KENNETH A. The collection in Trinidad and southern Brazil of coccinellids predatory on bamboo scales. Sixth Pacific Sci. Cong. held July 1939.
- BARTLETT, KENNETH A. Insectos beneficiosos: su intercambio entre los Estados Unidos y los países de Hispanoamérica. Hacienda 34: 428-430, illus. 1939.
- GARCÍA DE ARRILLAGA, NOEMÍ. Proyecto de perfumes de la Estación Experimental Federal de Mayaguez. Rev. de Agr. de Puerto Rico 31: 496-504, illus. 1939.
- PLANK, H. K., and SMITH, M. R. A survey of the pineapple mealybug in Puerto Rico and preliminary studies of its control. Puerto Rico Univ. Jour. Agr. 24: 49-76, illus. 1940.
- RUTTLE, MABEL L. Colchicine and the production of the new varieties of plants. Rev. de Agr. de Puerto Rico 31: 623-631, illus. 1939.
- WATSON, ALFRED N., and DAVIS, R. L. The statistical analysis of a spacing experiment with sweet corn. Amer. Soc. Agron. Jour. 30: 10-17, illus. 1938.

Annual Report was issued in English and Spanish.

The Annual Report for 1938, containing 137 pages and 36 illustrations, appeared in November 1939, and the reports for 1937 and 1938 were translated into Spanish and submitted for printing during the fiscal year 1940.

Interest taken in the work of the station by local Spanish-speaking farmers has been considerably stimulated by the publication of these reports in Spanish; during the past year requests for the Spanish edition were received from 324 individuals residing in Puerto Rico.

The Spanish edition also served to acquaint neighboring Western Hemisphere countries with the work the station is doing in tropical agriculture, the mailing list including 71 names with addresses in 13 such countries. This edition was also sent to 1 address in each of 3 other foreign countries, namely, Italy, Spain, and Portugal, and in the Philippine Islands.

CHANGES IN PERSONNEL

An increase in chemistry personnel was made possible.

Because of increased responsibilities, Congress approved an increase in the appropriation of the experiment station under which several new appointments have been made. Carlos Raoul Saavedra, junior agronomist, working under the Insular appropriation, resigned, effective September 2, 1939; his position was filled by the appointment of Antonio R. Villamil, who continued the activities of Mr. Saavedra in studies of the production of essential oils. Under the Insular appropriation Violeta Biaggi was appointed assistant librarian, effective September 2, 1939.

Miss Agnes Bechhold was appointed secretary to the director, effective September 5, 1939. The appointments of Merriam A. Jones to serve as junior chemist in studies of rotenone plants and their processing, and of Howard T. Love, junior chemist, for investigations of drug plants and their processing, were made effective November 1, 1939. M. Hollis Kannenberg was appointed clerk in charge of property records September 19, 1939. Beverly T. Taylor was appointed scientific aide to care for plant introductions and their propagation and distribution, effective September 14, 1939.

Wallace K. Bailey, associate horticulturist and acting director, was on leave effective October 14, 1939, to the end of the fiscal year.

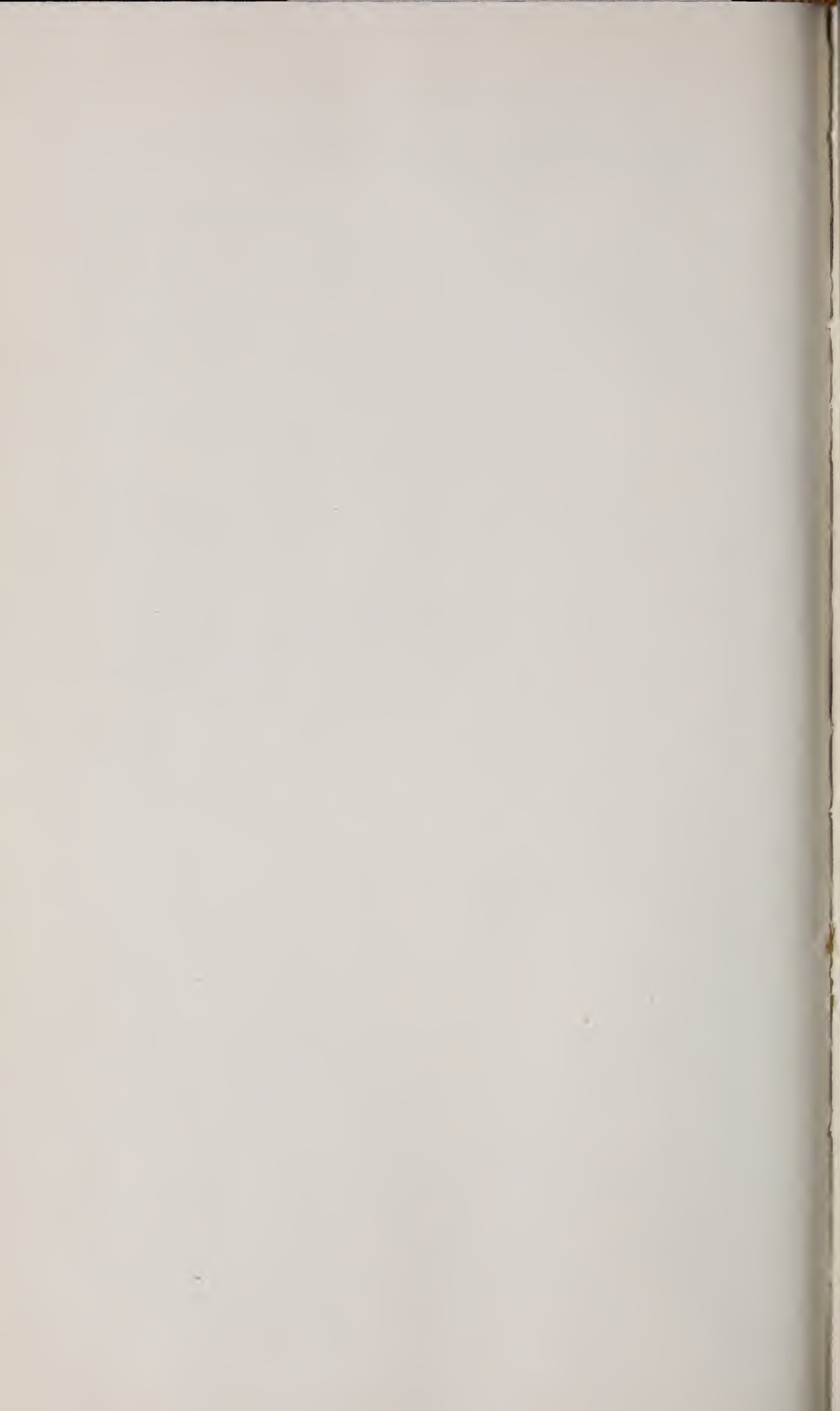
Personnel aided in development of Western Hemisphere agricultural policies.

During the year Atherton Lee, director, was detailed to the Office of Foreign Agricultural Relations for service for the Government of Ecuador during the months of November and December 1939, and for the Government of Colombia during January, February, and March 1940. In June his services were again loaned to the Office of Foreign Agricultural Relations to aid the agricultural work of the European refugee settlement in Santo Domingo. During the absence of the director on these assignments, Kenneth A. Bartlett served as acting director of the station.

LITERATURE CITED

- (1) ARBENZ, E.
1917. UEBER QUANTITATIVE BESTIMMUNG DER OXALSÄURE IN NÄHRUNGS- UND GENUSSMITTELN. Mitt. aus dem Geb. der Lebensmitl. Untersuch. u. Hyg. 8: 98-104.
- (2) BAILEY, WALLACE K.
1940. EXPERIMENTS IN CONTROLLING CORN EAR PESTS IN PUERTO RICO. Puerto Rico Agr. Expt. Sta. Cir. 23, 23 pp., illus.
- (3) BALLS, A. K., and HALE, W. S.
1933. DETERMINATION OF PEROXIDASE IN AGRICULTURAL PRODUCTS. Assoc. Off. Agr. Chem. Jour. 16: 445-453.
- (4) ——— and HALE, W. S.
1934. ON PEROXIDASE. Jour. Biol. Chem. 107: 767-782, illus.
- (5) BATEMAN, E.
1911. A VISUAL METHOD FOR DETERMINING THE PENETRATION OF INORGANIC SALTS IN TREATED WOOD. U. S. Forest Serv. Cir. 190, 5 pp., illus.
- (6) CHACE, ED. MACKAY, TOLMAN, L. M., and MUNSON, L. S.
1904. CHEMICAL COMPOSITION OF SOME TROPICAL FRUITS AND THEIR PRODUCTS. I. A STUDY OF CUBAN FRUITS. II. THE COMPOSITION OF FRESH AND CANNED PINEAPPLES. U. S. Dept. Agr., Bur. Chem. Bul. 87, 38 pp.
- (7) HARLAND, SYDNEY CROSS.
1937. A NOTE ON TWO LARVAL PARASITES OF THE SUGAR-CANE MOTH-BORER IN SAO PAULO, BRAZIL. Trop. Agr. [Trinidad] 14: 280.
- (8) PARRY, ERNEST J.
1921. THE CHEMISTRY OF ESSENTIAL OILS AND ARTIFICIAL PERFUMES. Ed. 4, v. 1, 549 pp., illus. London.
- (9) ———
1922. THE CHEMISTRY OF ESSENTIAL OILS AND ARTIFICIAL PERFUMES. Ed. 4, v. 2, 365 pp. London.
- (10) ———
1925. PARRY'S CYCLOPAEDIA OF PERFUMERY: A HANDBOOK ON THE RAW MATERIALS USED BY THE PERFUMER. v. 1, 432 pp. London.
- (11) PRATT, D. S., and DEL ROSARIO, J. I.
1913. PHILIPPINE FRUITS: THEIR COMPOSITION AND CHARACTERISTICS. Philippine Jour. Sci., Sect. A, 8: 59-80, illus.
- (12) STAHL, A. L.
1935. COMPOSITION OF MISCELLANEOUS TROPICAL AND SUB-TROPICAL FLORIDA FRUITS. Fla. Agr. Expt. Sta. Bul. 283, 20 pp.
- (13) THOMPSON, ALICE R.
1915. THE COMPOSITION OF HAWAIIAN FRUITS AND NUTS. Hawaii Agr. Expt. Sta. Rpt. 1914: 62-73.
- (14) TUCKER, C. M.
1927. VANILLA ROOT ROT. Jour. Agr. Res. 35: 1121-1136, illus.
- (15) TUCKER, R. W. E.
1939. INTRODUCTION OF DRY AREA RACE OF METAGONYSTYLUM MINENSE INTO BARBADOS. Barbados Dept. Sci. and Agr., Agr. Jour. 8: [113]-131.

- (16) UNITED STATES PHARMACOPOEIAL CONVENTION, COMMITTEE OF REVISION.
1936. PHARMACOPOEIA OF THE UNITED STATES OF AMERICA. 11th decennial
rev., 676 pp., illus. Easton, Pa.
- (17) WEHMER, C.
1929-31. DIE PFLANZENSTOFFE, . . . PHANEROGAMEN. Ed. 2, v. 1, 640 pp.
Jena.
- (18) WINTON, ANDREW L., and WINTON, KATE BARBER.
1935. THE STRUCTURE AND COMPOSITION OF FOODS. II. VEGETABLES, LE-
GUMES, FRUITS. 904 pp., illus. New York and London.
- (19) WOLCOTT, GEORGE N.
1936. "INSECTAE BORINQUENSES," A REVISED ANNOTATED CHECK-LIST OF THE
INSECTS OF PUERTO RICO. With a host-plant index by José I. Otero.
Puerto Rico Univ. Jour. Agr. 20: 1-627, illus.





The rainfall chart on the outside rear cover introduces two new methods in graphic representation of rainfall distribution. For the purpose of showing rainfall distribution, the year has been divided into 26 periods of 14 days each; this has the advantage over the usual separation of the year into 12 months in that the resulting curve is much more flexible and shows slight differences in rainfall trends during the year. Moreover, where the year is divided into 12 months February represents only 28 days of rainfall while March represents 31 days; in the graph as presented on the rear cover, each bar represents a uniform period of time, i. e., 14 days.

The graph also shows the standard deviation of rainfall from the mean and the absolute minimum rainfall in any one of the 14-day periods during the 28 years of record. With this graph, therefore, the agricultural producer in Mayaguez has a fairly complete knowledge of rainfall expectancy during the year.

